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INCREASED INTRACRANIAL PRESSURE

A Clinical Analysis of the Causes and Characteristics of Several Types

F. R. FORD, M.D., and E. L. MURPHY, M.D., M.R.C.P. Lond.

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Submitted for publication March 28, 1939

INTRODUCTION

The clinical neurologist must ever be on the alert to detect signs of increased intracranial pressure for this condition is of the greatest importance in the diagnosis and prognosis of disease of the nervous system. Unfortunately these signs are not always easy to interpret and it is frequently a matter of the utmost difficulty to form a correct estimate of the level of intracranial tension. Our uncertainty is largely due to the fact that increased intracranial pressure may be a result of a large variety of diseases and may be produced by several mechanisms, each of which presents certain individual characteristics. There are also a number of compensatory mechanisms which are adapted for the relief of increased intracranial pressure and these play significant rôles in determining the clinical picture. We propose to describe six different types of increased intracranial pressure and to analyse their clinical characteristics. Representative cases are cited but no attempt will be made to mention all conditions which may cause increased intracranial pressure. The following outline is offered:

Types of Increased Intracranial Pressure

1. Type due to Hydrocephalus
   - Obstructive
     - Congenital Defect
     - Neoplasm
     - Inflammatory
   - Excess of Secretion
     - Hypertrophy of the Choroid Plexus

2. Type due to Mass within the Cranium
   - Neoplasm
   - Abscess
   - Hematoma
   - Granuloma
   - Cyst
3. Type due to Swelling of the Brain
   - Trauma
   - Thrombosis of the Dural Sinuses
   - Hypertensive States, *e.g.*, acute glomerular nephritis, lead poisoning, *etc.*
   - Degenerative Processes, *e.g.*, Schilder's Disease.
4. Type due to extra-Cranial Obstruction to the Venous Return from the Cranium. *e.g.*, Failure of the right side of the heart, and mediastinal tumors.
5. Type due to Distention of the Intracranial Vascular Channels. *e.g.*, Arteriovenous fistula
6. Type due to Diminished Capacity of the Cranium. *e.g.*, Oxycephaly

Estimation of Intracranial Pressure: It seems to be generally accepted that the spinal fluid pressure of normal individuals in the lateral recumbent position varies between extremes of 100 and 200 millimeters of water when measured by a manometer attached to a hollow needle in the lumbar subarachnoid space. Pressures of over 200 millimeters are regarded as definitely abnormal and pressures between 180 and 200 millimeters are at least suggestive of increased intracranial pressure. It has been found that in certain pathological states involving the brain, as for example intracranial neoplasms, the intracranial pressure may exceed 600 millimeters of water. Estimation of pressure by such means permits exact measurements and has therefore obvious advantages. It is, however, attended by dangers and in some circumstances yields misleading results. It is of course well known that spinal puncture in cases in which the intracranial pressure is greatly elevated may cause collapse and even sudden death as a result of herniation of the medulla into the foramen magnum when fluid is withdrawn below.[1] Intracranial pressure is elevated significantly by muscular tension, fright, straining, weeping, coughing, holding the breath and pressure on the abdomen. In dealing with children and with confused or terrified adults it is often impossible to obtain correct readings. Hodgson found in studies made by combined ventricular and spinal punctures that in patients with subtentorial tumors communication between the ventricular system and the spinal subarachnoid space was obstructed in about two-thirds of

[1] Schaller claims that there is no proof that there is sudden wedging of the medulla into the foramen magnum as a result of spinal puncture. He gives reasons to show that the herniation is due to gradually developing edema of the brain which is in some way induced by the release of fluid. No doubt this is correct in many cases, but it cannot explain the deaths which occur within a few moments. (Schaller, W. F.: Propriety of Diagnostic Lumbar Puncture in Intracranial Hypertension. *J. Neurol. & Psychopath.* 1933, **14:** 116.)

all cases and in these the spinal pressure readings were apt to be lower than the intracranial pressure. This source of error is eliminated if one relies upon ventricular pressure readings, but this method necessitates trephining of the skull and puncture of the brain, procedures which demand not only the most careful asepsis but a proper degree of surgical skill. If general anesthesia is employed there may be some degree of asphyxia which causes elevation of intracranial pressure and if local anesthesia is used, fright and muscular tension may produce the same result. There is good reason to believe, moreover, that when the intracranial pressure is greatly elevated it is subject to spontaneous variations. Certainly the fluctuations in pulse, temperature and respiration which are observed in such cases suggest variations in intracranial pressure. The observations of Adson and Lillie are of particular significance in this connection. These investigators fixed a hollow needle in the lateral ventricle of a patient suffering from a glioma of the brain and made continuous readings of the pulse, respiration, blood pressure and intracranial pressure at hourly intervals for four days. It was discovered that the intracranial pressure was constantly changing and varied between extremes of 130 and 980 millimeters of water. There was sometimes a difference of 500 millimeters between two readings taken an hour apart. Ayer states that in six cases of brain tumor in which there was definite papilledema and the usual evidences of increased intracranial pressure he obtained low readings in ventricular pressure estimations. In some cases of this type the ventricular pressure may be actually below normal, possibly as a result of vomiting to the point of dehydration. It would seem correct to say, therefore, that one determination of intracranial pressure in a case of disease of the nervous system is no more instructive than one determination of the patient's blood pressure in hypertension. Ayer, whose careful investigations enable him to speak with authority, summed up the value of manometric studies of the spinal fluid pressure by saying, "At best, then one usually learns what is probably already apparent from a fundus examination."

It is possible in most cases to estimate with some degree of accuracy the level of the intracranial pressure by study of the physical signs and symptoms. In cases in which the pressure rises slowly, headache is almost invariably the initial symptom. This may be continuous but is more often intermittent or paroxysmal. Vomiting usually appears later and probably indicates a higher degree of pressure. Papilledema is the next sign to appear. This demands not only pressure of a certain degree but has a certain latency so that even intense pressure will not cause papilledema until several days have elapsed. For a time the degree of elevation of the optic discs may be roughly correlated with the height of the pressure. Eventually, however, the swelling of the discs will recede even when the pressure remains high. Diplopia may follow or precede papilledema. At first it is intermittent but later may become constant. At this point there is some drowsiness and slight clouding of consciousness. Later the drowsiness deepens into stupor which passes slowly into coma. Deep stupor or coma is apt to be associated with general muscular rigidity, retraction of the head, difficulty in swallowing, slowing of the pulse and respiratory rates, dilatation of the pupils and loss of their light

reflexes. Such signs indicate an extremely high pressure. At this stage a systolic bruit may be heard in the occipital region. Finally the pulse and respiratory rhythms become irregular and death results from respiratory failure.

The signs and symptoms mentioned above are, of course, results of generalized increase of intracranial pressure but all of them are really focal signs and each demands special consideration. The cause of the headache is not entirely clear, but since it is found that during operations under local anesthesia, the only procedures which cause pain are those which traumatize the meninges, it may be assumed that stretching or compression of the meninges is responsible. The sensory nerves in the meninges seem to be closely associated with the blood vessels. More sharply localized pains may be due to compression of sensory nerves at the base. Diplopia is usually due to paresis or paralysis of one or both sixth nerves. Many years ago Cushing showed that this is most frequently due to compression of the nerve by one of the transverse branches of the basilar artery when the pons is forced down upon the base of the posterior fossa. The vomiting, bradycardia, difficulty in swallowing and the slowing and eventual failure of respiration may all be explained by herniation of the medulla into the foramen magnum. Jefferson has recently discussed the significance of tentorial herniation. He suggests that crowding of the temporal lobe into the incisura as a result of pressure from above causes general muscular rigidity due to transmitted pressure on the crura and dilatation of the pupils with loss of light reflex due to pressure on the third nerves. The disturbances of consciousness result, no doubt, from compression of the cerebral cortex and reduction of its circulation. The cause of papilledema has been a favorite subject of debate. Suffice it to say that it is now accepted by most authorities that papilledema is a result of distention of the optic nerve sheath with cerebrospinal fluid under pressure so that the central retinal vein, which lies within the distal third of the nerve, is compressed and any lymphatic channels which may exist in the nerve are likewise obstructed. If the optic nerve is compressed directly by a neoplasm or if its sheath is occluded by an inflammatory process so that cerebrospinal fluid cannot flow into its distal part, papilledema will not occur even when the intracranial pressure is very high.[2] The systolic bruit in the occipital region is attributed to herniation of the medulla into the foramen magnum with compression of the vertebral artery against the bony margin in such a way as to cause narrowing of its lumen.

It may be well at this point to state that elevation of intracranial pressure rarely causes any change in blood pressure. Only in certain cases of sudden and intense rise in intracranial pressure, as for example in rupture of the middle meningeal artery, is this effect clearly demonstrable. Merritt states that arterial hypertension is scarcely ever produced by intracranial tumors and that it is only in cases

[2] It has been claimed that the level of intraocular pressure has a definite influence upon the development of papilledema. If there is gross inequality of pressure in the two eyes, papilledema will develop sooner in the eye in which the tension is low.

in which the intracranial pressure rises to approximately the level of the arterial pressure that the latter shows a compensatory elevation.

Cases of increased intracranial pressure resulting from congestive heart failure or obstruction of the venous drainage of the head due to tumors of the mediastinum, seem to constitute exceptions to the general rule for it is claimed that they are not associated with any of the signs or symptoms commonly found in increased intracranial pressure due to other causes.

TYPE DUE TO HYDROCEPHALUS

A. Obstructive

Hydrocephalus is the commonest cause of increased intracranial pressure. It is almost invariably a result of obstruction of the ventricular system or of the subarachnoid spaces through which the cerebrospinal fluid flows. When the normal circulation is impeded the fluid cannot be absorbed and accumulates under increased pressure causing dilatation of the ventricles with compression and atrophy of the cerebral cortex. The elevation of intracranial pressure would seem to be determined by the pressure under which the cerebrospinal fluid is secreted.

Acute Hydrocephalus: The onset of symptoms is often very abrupt even in cases in which the obstruction is due to slowly growing neoplasms. Headache and vomiting are followed within a short time by drowsiness and bradycardia. Internal squints due to sixth nerve palsies may occur, and within a few days there is papilledema. Soon the patient becomes stuporous and finally passes into coma associated with general muscular rigidity. The pulse and respiration become irregular and finally death occurs from failure of respiration. The whole course of events may occur within a few days or hours.

Compensated Hydrocephalus: The above discussion applies to acute uncompensated hydrocephalus. In most cases of sudden occlusion of the ventricular system the course is not so brief and there is evidence of compensatory mechanisms. As a rule if the patient survives more than a short time there is gradual reduction of intracranial pressure even if no therapeutic measures are employed. The changes in the pulse and respiratory rates disappear, vomiting ceases and the state of consciousness improves. Papilledema recedes, leaving some degree of optic atrophy and reduction of vision. Disorders indicative of injury to the hypothalamus may be explained by distention of the third

ventricle which is also a possible cause of damage to the optic chiasm with consequent optic atrophy. In some cases mild spasticity is found which is to be attributed to atrophy of the motor cortex. Observations made upon patients who have had subtemporal decompression performed make it clear that the intracranial pressure may eventually become quite normal even when the ventricular system is still completely obstructed. Such compensation is gradually acquired during a period of months. In most cases the intracranial pressure never falls to the normal level but remains somewhat elevated.

The explanation of this compensation is not entirely clear. Wislocki and Putnam have shown that there is some absorption of fluid through the ependyma in obstructive hydrocephalus though absorption by this route is never adequate. The dehydration resulting from vomiting must play a rôle in compensation when pressure is very high, and the compression of the cortex and dilatation of the ventricles may also act as a safety valve. It seems necessary to assume, however, that there is another element in compensation and we are inclined to believe that there is actually diminution of secretion of cerebrospinal fluid in long standing cases. If one inspects the choriod plexuses in cases of chronic hydrocephalus, it is seen that they are small and flattened. Hassin emphasizes this appearance and claims that atrophy of the plexuses is the cause of hydrocephalus for he thinks that their function under normal conditions is the absorption of cerebrospinal fluid. We are inclined to believe, on the contrary, that the compression and apparent atrophy of the choroid plexuses is a result of hydrocephalus, not a cause, and that it is the major factor in compensation. The plexuses may be compressed by the intraventricular pressure to such an extent that their blood supply is diminished and their secretory activity reduced to a minimum. If ventricular puncture is performed in a case where compensation has been established and some fluid is removed, there may be a great rise in pressure within a few hours with all the signs of acute hydrocephalus. We are inclined to believe that the sudden withdrawal of fluid causes hyperemia of the plexuses with prompt outpouring of large amounts of fluid. The reaction is comparable to that of the kidney when an obstruction of the ureter has been removed. This unfortunate train of events is illustrated in the following case history:

Case History (Path. 7643). Girl of 24 years developed headache and papilledema. Decompression. Disappearance of all signs of increased intracranial pressure. Two years later ventriculogram revealed tumor in third ventricle. After this procedure intracranial pressure rose again and ventricular punctures were required at 6 hour intervals until operation a month later. Death. Post-mortem examination revealed glioma causing complete obstruction of the third ventricle and hydrocephalus.

H. P., a previously healthy girl of 24 years began to have severe headaches in April 1921. During the more severe headaches she would have double vision and would often vomit. In some attacks she would grow weak and dizzy. These symptoms became more severe and frequent. In August she was taken to a neurosurgeon who found bilateral papilledema but could not localize the lesion. In the hope of relieving the pressure he performed subtemporal decompression on the right side. For a time this decompression bulged over the right ear, but later it became flat and the papilledema disappeared. The patient still had attacks of weakness and dizziness, so she came to Dr. Dandy on July 12, 1923.

Examination revealed a well nourished and well developed young woman of the given age, who was mentally clear and in no distress. The area of the decompression was soft and sunken. The optic nerves were slightly pale and the disc margins were indistinct as in secondary atrophy, but central vision was good and the visual fields showed only slight concentric constriction. Neurological examination was otherwise negative. Ventriculography was performed on July 14. Some 250 cc. of fluid were removed and replaced by air. Study of the roentgenograms revealed marked hydrocephalus of the lateral ventricles and a filling defect in the third ventricle which was taken to indicate a tumor. It was decided that the tumor was probably inoperable. After the ventriculogram the patient developed signs of acute hydrocephalus and ventricular puncture had to be repeated every 6 hours. A few hours after each puncture the patient would develop headache, vomiting, stupor, muscular rigidity, dilatation of the pupils, alarming changes in the pulse and temperature with bulging of the decompression so that another puncture was required. This unfortunate situation persisted until August 22 when a desperate effort was made to remove the tumor or drain the ventricles. The patient failed to rally and died on August the 24th. Post-mortem examination revealed an infiltrating glioma filling the third ventricle and extending into the aqueduct. The ventricular system was completely occluded.

The rôle of the decompression in such cases would seem to be merely that of a palliative procedure which is of value during the period in which compensation is being established. The normal adult skull behaves, as Weed has shown, almost as if it were a rigid walled container or a "closed box," as he phrases it. The slightest increase in fluid content of such a system causes at once an enormous rise in pressure. Perhaps this physical property of the cranium is an important factor in producing the extreme variations in intracranial

pressure observed by Adson and Lillie. Subtemporal decompression replaces a part of this rigid wall by soft tissue which possesses a degree of elasticity and thereby prevents extreme variations in pressure, but it cannot have any influence in reducing pressure to a normal level, for any increase in the capacity of the cranium which may result, is at once taken up by an increased amount of cerebrospinal fluid.

Occasionally one sees cases of compensated hydrocephalus in which there is no history of the acute symptoms which usually mark the onset of obstruction. Riddoch has recently reported two cases of this type with a very interesting discussion. Despite the presence of obstruction of the third ventricle and of hydrocephalus, there was no papilledema and no optic atrophy. Indeed, there was no evidence that the patients had ever had high grade increased intracranial pressure. In such cases one is forced to assume that the obstruction has developed so gradually that compensation has been able to keep pace with it.

Congenital hydrocephalus offers the commonest examples of compensated hydrocephalus, however. The onset is almost certainly during intrauterine life, for the anatomical findings are such that one is justified in assuming that the cerebrospinal fluid channels were never properly formed. Moreover, the child's head begins to grow at an abnormal rate immediately after birth. The power of compensation seems to vary within wide limits in these cases. Sometimes the head continues to enlarge until it reaches an enormous size and the cortex is reduced to a thin translucent membrane. In other cases the process becomes arrested at an earlier stage and the patient may live to an advanced age with a large head but without any other evidence of increased intracranial pressure. During infancy there is never papilledema or other evidence of high grade pressure and manometric estimations of the intraventricular pressure give readings barely above the normal. No doubt this is due to the elasticity of the infantile skull which is merely a membranous sac with bony plates in the walls and is quite unlike the rigid skull of an adult.

Failure of Compensation: In some cases compensation fails for no apparent reason. This may occur at any age and we have observed it at the ages of eight, nine, thirteen and forty-nine years. In such cases, a patient who has had an enlarged head since infancy, but no

headaches, vomiting or other signs of increased intracranial pressure develops papilledema and the usual evidences of acute hydrocephalus. We have no explanation to offer for this failure of compensation unless one might wish to assume that a partial obstruction has finally become complete. The lesions found at post-mortem examination are merely those usually found in congenital hydrocephalus. The following case history illustrates these statements:

Case History (U-14357. Path. 10152). Girl whose head was large at birth and enlarged rapidly during infancy. Later no signs of increased pressure but at the age of 8 years signs of acute hydrocephalus developed with papilledema and sixth nerve palsy. Operation revealed imperfect formation of foramina of the 4th ventricle. Death. Post-mortem examination.

M. W. L. was born by a difficult forceps delivery. Her head was thought to be abnormally large at birth and grew too rapidly for a few months. After this the growth of the head was normal. There was never any evidence of headache and the child rarely vomited. She began to talk at the age of 9 months but was slow in learning to walk. Her gait was slightly unsteady and she fell more frequently than the other children. She was also clumsy in the use of her arms. However, the child was vigorous and enjoyed excellent health. Her parents, who believed in routine examinations, were assured by several pediatricians that the child was quite normal in every way.

On September 7, 1927 at the age of 8 years, the child complained of severe headache and vomited several times. The headache persisted and a few days later the child was found to have diplopia and an internal squint of the left eye. At the end of a week the headache which had been constant at first was becoming intermittent and the child was growing drowsy, especially during the paroxysms of pain.

On September 22 the child was brought to Dr. Dandy. She was a well developed and well nourished little girl who was semistuporous at times. Her head was slightly larger than normal and of hydrocephalic shape. There was bilateral papilledema of 3 diopters which appeared to be quite fresh and was not associated with any optic atrophy. The left external rectus was weak, and double vision was present. Mild ataxia (which seemed to be of cerebellar type) was found in all four extremities, and there was a general increase in the tendon reflexes. The plantar responses were normal. All other examinations were negative.

Exploration of the posterior fossa revealed that the foramina of the 4th ventricle were occluded and that there was pronounced hydrocephalus. The cisternae about the medulla were obliterated and the cerebellum and medulla were adherent to the arachnoid. Dr. Dandy studied these appearances carefully and decided that the foramina of Luschka had never been patent and that the foramen of Majendie had been patent until recently but had closed as a result of proliferation of connective tissue about the opening. The whole condition was regarded as a

failure of the subarachnoid spaces to develop properly. Despite repeated efforts, it was impossible to keep the foramina open and the child eventually died. Post-mortem examination confirmed the findings described at operation.

B. Excess of Secretion

Little can be said about this condition for only a few cases are on record in which one can be confident that obstruction played no part in the development of hydrocephalus. These cases are described under the title of hypertrophy of the choroid plexus and their clinical characteristics are apparently identical with those of the common congenital obstructive hydrocephalus of infants. L. E. Davis has described a case of this type. Evidently the intracranial pressure may be greatly elevated by the excessive secretion of cerebrospinal fluid which is poured out in such abundance that even a normal absorptive mechanism cannot keep pace with it. The cause of such hypertrophy is quite unknown. A case of our own may be cited to illustrate this condition:

Case History (Path. 14946. U-72729). Child of 5 months developed rapidly progressing hydrocephalus. Operation disclosed bilateral hypertrophy of the choroid plexuses of the lateral ventricles. No obstruction found at post-mortem examination.

I. C. was born at full term by normal labor. He seemed to be quite healthy until the age of 5 months when his head began to grow at an abnormal rate. He soon became unable to hold up his head, and lost the other motor reactions he had acquired.

Examination on September 11, 1936 when the child was 6 months old, revealed a typically hydrocephalic head which measured 45.5 cm. in the occipitofrontal circumference. The fontanels were abnormally wide and tense. Neurological examination was negative. Dye injected into the spinal subarachnoid space was recovered promptly from both lateral ventricles and was excreted by the kidneys just as rapidly as in a normal baby, showing apparently that there was no obstruction and no defect in absorption.

Operation disclosed great masses of choroid plexus in both lateral ventricles. These were removed as completely as possible but hemorrhage occurred and the child died. Post-mortem examination revealed that the entire ventricular system including the aqueduct and the fourth ventricle was uniformly dilated. There was no obstruction in any part of the system. The choroid plexuses were greatly hypertrophied and large enough to fill ventricles of normal size.

TYPE DUE TO A MASS WITHIN THE CRANIUM

Most brain tumors cause increased intracranial pressure because they produce obstruction of the ventricular system and hydrocephalus,

but occasionally one finds high grade increased intracranial pressure in cases of tumor where the ventricular system is not obstructed. At operation or post-mortem examination one is sometimes astonished to see what an enormous mass may form in the cranium before death results and to observe how tolerant the brain is of slowly developing compression. All tumors seem to be alike in this respect, whether they are neoplasms, cysts, hematomas, granulomas or encapsulated abscesses. The chronic subdural hematoma is probably the best example of a tumor which behaves in this fashion because it compresses the brain to an extraordinary extent and does not cause hydrocephalus. We shall discuss subdural hematoma as representative of this group.

The first symptom is almost invariably headache. One gains the impression that this is not very intense for the patient usually mentions it rather casually. The only other constant sign is deepening drowsiness which during a period of days or weeks passes slowly into stupor and finally into coma. Papilledema occurs in only a small percentage of cases and in these it is mild and appears late. Vomiting and palsy of the sixth nerve are uncommon. Bradycardia has been described as an important sign in subdural hematoma but in our experience develops late if at all. Focal signs are notoriously unreliable. Hemiparesis may be found but this again is a late and inconstant sign. It may be homolateral or contralateral to the hematoma. Dilatation of the homolateral pupil is often seen but may be absent throughout the entire course.

At operation or post-mortem examination one finds a large hematoma overlying the lateral aspect of the hemisphere. It may extend from the frontal to the occipital pole and lies as a rule entirely above the fissure of Sylvius, being thickest near the vertex. The homolateral hemisphere is compressed so that its ventricle may be displaced beyond the midline to the opposite side. An interesting feature of many cases is the striking edema of the homolateral hemisphere which probably plays a rôle in the production of symptoms. The ventricular system is not dilated in any part and we may assume that it is not obstructed.

The explanation of this picture is not easy. The absence of papilledema, palsy of the sixth nerve, bradycardia and vomiting would suggest that we do not have a marked elevation of intracranial pressure, incredible as it may seem. This statement is supported with

spinal fluid pressure readings by Gardner and others, which show that pressures over 200 millimeters of water are found in only about half of these cases. The brain evidently yields so readily to slow local pressure that high grade increase of general intracranial pressure does not occur until an extraordinary degree of compression has been produced. If one reflects that the brain is quite soft and plastic and that the pressure within the ventricles and in the cerebral veins is very low, it is easy to understand how this is possible. Very slight pressure is sufficient to squeeze fluid out of the cerebral sulci, ventricles and tissue spaces and to reduce the volume of blood in the veins. Dandy has illuminated this process in his discussion of "The Space-compensating Function of the Cerebrospinal Fluid." No doubt the progressive reduction of the level of consciousness which is such a characteristic feature of these cases is due to compression of the brain. The focal signs are produced indirectly. Kernohan and Woltman explain the homolateral hemiparesis by indentation of the contralateral crus by the margin of the tentorium as a result of displacement of the brain. Jefferson has shown, in an interesting paper mentioned above, that the contralateral hemiparesis and dilatation of the homolateral pupil are due to herniation of the temporal lobe through the tentorium with compression of the homolateral aspect of the midbrain.

TYPE DUE TO SWELLING OF THE BRAIN

In this group we include a number of heterogeneous conditions resulting from venous obstruction, intoxications, trauma and degeneration which have only one feature in common, that is, they are associated with swelling or edema of the brain which may in some instances give rise to increase of intracranial pressure.

Traumatic Lesions of the Brain: It is generally believed that in acute traumatic lesions of the brain, after the phenomena due to shock have passed off, the chief signs and symptoms are due to increased intracranial pressure and that the outcome depends upon the degree of this pressure and the surgeon's success in combating it. This rise in pressure is attributed to edema of the brain and intracranial hemorrhage. The studies of Dandy and of Woodhall have revealed how the changes in intracranial pressure are indicated by the variations in pulse, respiration, temperature and the state of consciousness. When

these signs indicate steadily mounting pressure the surgeon performs subtemporal decompression. When the dura is incised, the brain often herniates under pressure and in some cases the protrusion of the brain is so violent that the cortex may rupture unless extreme care is used. At post-mortem examination it is usually stated that the brain is swollen and edematous and may or may not show areas of hemorrhage and necrosis. Courville who has made a very careful and detailed study of the brain in nearly 500 fatal head injuries, states:

"While this condition (edema) occurs almost invariably after any injury to the head, it is often limited in degree and exerts no discernible clinical effect. In those instances, however, where it is more extensive, two types may be distinguished; (a) general edema and (b) localized edema—. In the writer's opinion, it is the severe localized edema accompanying gross severe contusions of the brain that is most intractable and most likely to result in the patient's death."

In recent years several studies of the spinal fluid pressure in cases of acute cerebral injuries have been made and it is stated that definite elevation of intracranial pressure is unusual and that when the pressure is elevated it is rarely above 300 millimeters of water. It is claimed, moreover, that the level of intracranial pressure does not determine the state of consciousness and does not cause death in fatal cases. Russell suggests that in fatal cases, "immediate and delayed shock" play the most important rôles. Zierold states that death must be due to "disturbance in the medullary part of the brain" and suggests that this disturbance may be "localized pressure." Apparently by localized pressure he means localized edema of the medulla. To support their pressure readings these investigators emphasize the fact that papilledema is rare after head injuries.

It is not easy to reconcile these conflicting views. It may be regarded as established that the vast majority of cerebral traumas cause some degree of edema and that severe contusions cause gross, localized edema. The striking tendency of the brain to herniate at operation unquestionably indicates that there is increased intracranial pressure in many cases. On the other hand, the studies of spinal fluid pressure mentioned above would seem to indicate that increased intracranial pressure is unusual. We have pointed out above that measurement of the spinal fluid pressure may be misleading at times

but one can scarcely disregard a large series of fairly consistent observations by several independent investigators. It is possible that the local edema may be compensated by compression of the more normal parts of the brain and squeezing out of fluid from the sulci and the ventricular system, so that a general increase of intracranial pressure does not occur until this compensation has reached its limit. No doubt the progressive reduction of consciousness (in many cases) may be due to edema and compression of the brain and not to rising pressure. It is also possible that some of the changes in the pulse, respiration and temperature which are taken to indicate increased intracranial pressure may be due to developing edema of the medulla so that increased intracranial pressure, although it unquestionably occurs in many cases, is a less important cause of death than it is generally believed to be.

Thrombosis of the Dural Sinuses: A number of important studies have been made of this subject in recent years by Woodhall, Symonds, Byers and Hass which have revealed among other facts that occlusion of the dural sinuses is a more important cause of increased intracranial pressure than had been realized in the past. The essence of the problem is really very simple. If the thrombus is limited to one sinus, it is not likely to cause much disturbance of the circulation since the abundance of collateral channels provides ample drainage. If, however, the thrombus extends into the collaterals, as it frequently does, there may be complete obstruction of the venous drainage of large parts of the brain with the development of massive edema and of hemorrhages. Increased intracranial pressure frequently results. Woodhall has pointed out that there is a great deal of variation in the anatomy of the dural sinuses and that such variations are of great importance in determining the effect of occlusion in any part of the system. The following case history is illustrative of the effects of extensive dural sinus thrombosis.

Case History (U-20828. Path. 10607). Woman of 26 years developed headache, vomiting and papilledema. Ventriculography revealed small, symmetrical ventricles. Decompression. Larger opening in skull demanded by rising pressure. Rupture of wound with herniation of brain. Resection of temporal lobe required to close wound. Death. Post-mortem examination. Thrombosis of the superior longitudinal and left lateral sinuses.

J. P., a previously healthy young woman of 26 years, developed a sudden and severe headache on September 12, 1928. This headache persisted and during the next few days she developed diplopia, began to vomit and finally became stuporous. There were no focal signs, and no other significant facts were elicited.

Examination on September 24 revealed a well developed and well nourished girl, who was semi-stuporous and evidently in severe pain. The optic discs were edematous and there was weakness of both sixth nerves. Neurological examination was otherwise negative except for the fact that Babinski's sign was positive on both sides. All other medical and laboratory examinations were negative.

Ventriculography was performed by Dr. Dandy on September 26. The ventricles were found to be small and symmetrical. The cerebrospinal fluid was under great pressure and was tinged with blood. In the next few days the stupor deepened and repeated spinal punctures were performed to reduce intracranial pressure. These measures were without avail and it was necessary to perform a right subtemporal decompression. The brain was greatly congested and herniated into the opening, so closure was difficult. The pressure was not satisfactorily relieved and the area of the decompression was very tense and bulging. Large quantities of fluid under high pressure were removed by spinal puncture in the next few days, but the patient sank into coma. On September 29, a large bone flap was removed in the hope of affording relief. Through this opening the brain herniated to an extraordinary degree. On October 6 the wound suddenly ruptured and the brain was exposed. The right temporal lobe had to be resected to secure closure, and the patient died in a few hours.

Post-mortem examination revealed extreme congestion and edema of the brain. There was a partially organized thrombus occupying the entire length of the superior longitudinal sinus with its collateral channels, the torcular Herophili and the left lateral sinus. The thrombi were of the non-septic type and it was impossible to discover their origin. The pathologist estimated their age at between two and three weeks.

Theoretically one might imagine several factors which contribute to the production of increased intracranial pressure in such cases. 1—Edema of the brain which is unquestionably present. 2—Hemorrhage which is present in the majority of cases, but not in all. 3—Distention of the intracranial venous channels which is always present to some degree. 4—Diminished absorption of cerebrospinal fluid as a result of obstruction of the veins into which the fluid is normally absorbed. This has been stressed by Symonds. 5—Increased production of cerebrospinal fluid due to congestion of the choroid plexuses. This factor can be regarded as merely a theoretical possibility. At post-mortem examination the brain is much swollen, congested and often studded by large or small hemorrhages. The ventricles are

small and compressed, the convolutions are flattened and the sulci narrowed.

If the patient survives the acute stages of this process, there is, as a rule, a gradual reduction of the intracranial pressure to a normal level in the course of a few weeks. This must be due to dilatation of collateral channels and later to recanalization of the thrombosed vessels. The following case history may serve to illustrate these features:

Case History (U-43253). Girl of 7 years developed left otitis media followed by headache, vomiting and papilledema. Cerebral ventricles small and spinal fluid normal. Subtemporal decompression performed. Gradual disappearance of pressure signs. Two years later mastoiditis, meningitis and death. Post-mortem examination revealed that left lateral sinus was replaced by fibrous cord due to organization of old thrombus.

B. H., a previously healthy girl of 7 years, developed otitis media on the left, Christmas, 1931. About a week later the child complained of headache and began to vomit. Vision became impaired and it was noticed that the left eye turned in.

Examination on January 28, 1932 revealed well developed papilledema and palsy of the left sixth nerve. The neurological examination was otherwise negative, and there was no active otitis or mastoiditis. Ventriculography revealed small but symmetrical ventricles. The spinal fluid was under pressure but otherwise normal. Dr. Dandy performed a subtemporal decompression on the right. The brain was under great tension and an excess of fluid was found in the subarachnoid spaces. For some weeks the decompression bulged but later the papilledema receded and the decompression became flat. The child was then quite well for two years.

In February, 1934 the patient once more developed otitis on the left. Signs of mastoiditis soon appeared and despite operation, meningitis developed. Death occurred after a short illness. At post-mortem examination, in addition to the evidences of a fresh meningitis, it was found that the left lateral sinus was replaced by a fibrous cord with a few small vascular channels running through it. In the opinion of the pathologist these appearances indicated that the left lateral sinus had been thrombosed during the illness two years before and the thrombus had been organized and partially recanalized.

Hypertensive States: Blackfan and others have shown that there may be elevation of intracranial pressure in *acute glomerular nephritis* of childhood. Headache, vomiting, slowing of the pulse rate and elevation of the spinal fluid pressure to over 300 millimeters of water may occur. In infants the fontanels bulge and the sutures of the skull may be separated. This clinical picture is often termed hypertensive en-

cephalopathy. During such episodes the arterial blood pressure is always greatly elevated and it is emphasized by many writers that the cerebral symptoms are more apt to occur when the rise of blood pressure is sudden, and that they parallel approximately in severity the degree of hypertension. All writers agree that striking edema of the brain is found at post-mortem examination in such cases. The convolutions are flattened, the ventricles compressed and a typical pressure cone is found at the base of the cerebellum. The brain may weigh 20 to 30 per cent more than normal. The increase of intracranial pressure is regarded as a direct result of the cerebral edema but the cause of the edema is not clear. There is not necessarily any significant increase of the non-protein nitrogen of the blood, and the 'phthalein excretion may be within normal limits.

Lead encephalopathy is another condition which is associated with swelling of the brain and increased intracranial pressure. The mechanism by which this is brought about is still unknown but it is constantly associated with arterial hypertension and is, in this respect, comparable to what is observed in acute glomerular nephritis. Blackman has shown that the brain, in lead poisoning, contains a great excess of coagulable fluid which lies in the interstices of the cells and which is no doubt responsible for the swelling. There is outspoken papilledema in some cases and the signs of increased pressure may be so severe that subtemporal decompression is necessary. In such cases the brain may herniate through the bony defect in the skull to an extreme degree and the herniation may persist for at least two months.

Case History (Ped. A-8487). Child of 3 years developed lead encephalopathy with papilledema and mental changes. Pulse between 60 and 70. Subtemporal decompression. Pronounced herniation of brain through bone defect for more than 2 months.[3]

S. K. was regarded as a normal child during infancy. He had always put things into his mouth which were unfit for food, and during the fall of 1938 he began to chew paint off the furniture. About the middle of September he began to vomit and showed signs of headache. He lost weight and seemed to be ill.

On October 3, at the age of 34 months, he was brought to the Harriet Lane Home. He was found to be pale and undernourished. At times he was restless and irritable and at other times apathetic and stuporous. A faint lead line was

[3] This case is presented by the courtesy of Dr. Edwards A. Park.

seen on the gums. There was well developed bilateral papilledema. Neurological examination revealed only a coarse tremor of the hands. There was a moderate secondary anemia and the red blood cells showed basophilic stippling. Roentgenograms of the long bones revealed typical lead lines, and spectroscopic study of the blood showed 0.39 mg. of lead per 100 cc. The spinal fluid was under greatly increased pressure and contained 10 cells per cu.mm. The Pandy test was strongly positive. During the next few days the temperature remained normal but the pulse rate varied between 60 and 70.

Right subtemporal decompression was performed. The brain was under great tension and herniated so strongly that closure was difficult. After operation the child's mental condition gradually improved, the pulse rate became normal and the papilledema receded. The brain herniated strongly through the defect in the bone. This herniation persisted unchanged through November and December. In January it was smaller and softer and in February it disappeared.

In *eclampsia gravidarum* one finds violent convulsions associated with arterial hypertension of high degree and abrupt onset. There may be headache, vomiting and other evidences of increased intracranial pressure. The brain is said to be edematous and it is believed that the edema is the cause of the increased intracranial pressure. Fishberg groups the cerebral symptoms of acute glomerular nephritis, lead encephalopathy and eclampsia together as examples of hypertensive encephalopathy and believes that they are connected in some manner with the arterial hypertension.

It has been shown by a number of investigators including Shelburne, Blain and O'Hare, Larsson, Fishberg, McAlpine and Grant that various types of *chronic arterial hypertension* of both renal and "essential" types in children and in adults may be associated with increased intracranial pressure. "Malignant" types of hypertension are especially apt to be complicated by increased intracranial tension. In such cases there is headache, vomiting and papilledema in addition to "albuminuric retinitis." The spinal fluid pressure is definitely elevated. In a study of 50 cases of hypertension Shelburne found spinal fluid pressure elevated in 20. Among these, 19 exhibited papilledema. Larsson gives similar figures. Pickering found that increased intracranial pressure is more apt to develop in young patients than in old and that it is of bad prognostic import. These authors conclude that in hypertension, papilledema is always indicative of increased intracranial pressure.

It is not clear how this elevation of intracranial pressure in hypertension is brought about. Since it is not found in all cases of severe hypertension it seems obvious that it is not directly related to the systolic arterial pressure. Pickering's investigations seem to show that there is some relation between diastolic pressure and the intracranial tension. Since the diastolic pressure approximates more closely the mean arterial tension, it is suggested that the latter pressure may determine the pressure of the cerebrospinal fluid. It is generally admitted that the level of intracranial pressure is not paralleled by the state of renal function for pressure may be normal in renal failure and may be elevated when no renal damage can be demonstrated. Shelburne and Pickering agree that elevation of venous pressure is not significant in the group of cases they investigated.

The anatomical investigations in these cases are still incomplete. One might expect to find edema of the brain, but in our experience the brains are usually small and shrunken as a result of vascular disturbances. Moreover, in the cases in which ventriculography has been performed to exclude tumor, we have found dilated ventricles rather than the small ventricles which one would expect in edema of the brain.

Case History (U-155654. Path. 16166). Man of 33 years with hypertension, headaches and bilateral papilledema of 3 diopters. Spinal fluid pressure 320 mm water. Ventriculogram normal. Death following abdominal sympathectomy. Post-mortem examination revealed no edema of the brain.

F. M., a man of 32 years, developed severe headaches and nocturia. After these symptoms had been present for several months, the patient's vision began to fail. He then came to the Johns Hopkins Hospital.

Examination revealed a well developed and well nourished young man who was well oriented and coöperative. Blood pressure was 240/160. There was bilateral papilledema of at least 3 diopters with flame shaped hemorrhages and fresh exudates about the discs. The retinal arteries were narrow. Vision was reduced to about 20/70 in both eyes. The fields were of normal outline but there was great enlargement of the blind spots. Neurological examination disclosed nothing of significance. The urine was negative. Various tests of renal function gave normal results. The spinal fluid was under pressure of 320 mm of water. Because of the possibility of brain tumor a ventriculogram was performed. This revealed ventricles of normal size. There was slight dilatation of the posterior horn of the right lateral ventricle but this was not regarded as important.

It seemed evident, therefore, that the patient's symptoms were due to hypertension and after some discussion it was decided to perform abdominal sympathectomy. This operation was carried out on the left side without difficulty. Several days later the same operation was performed on the right side. The patient was deeply shocked and died within a few hours.

Post-mortem examination revealed widespread arteriolar sclerosis and only moderate thickening of the larger arteries. The left renal artery was partially occluded by a large arterioslerotic plaque. The heart was only slightly enlarged. The brain was not edematous. The convolutions were rounded, the sulci of normal depth and there was no cerebellar pressure cone. The ventricles were of normal size. There was a slight dilatation of the posterior horn of the right lateral ventricle. Microscopic examination of the brain was negative.

Schilder's Disease: It is not unusual to discover swelling of the optic nerve heads in Schilder's disease and in various other demyelinizing processes affecting the central nervous system. In most instances, however, this swelling of the discs is due to optic neuritis and does not indicate elevation of intracranial pressure. In a small percentage of cases of Schilder's diseases there is true papilledema due to increased intracranial pressure. In our experience, this is due to edema of the cerebral hemispheres in association with massive and acute lesions. In the case cited below, the patient's death was directly due to increased intracranial pressure.

Case History (U-11887. Path. 9830). Boy of 14 years developed headache, vomiting, internal squint, weakness and spasticity of both legs and left arm, and bilateral papilledema in course of 6 weeks. Death resulted from increased intracranial pressure. Post-mortem examination revealed encephalitis periaxialis diffusa with great swelling of the right hemisphere.

O. W., a boy of 14 years, developed headaches and vomiting about March 15, 1927. Diplopia soon appeared and was followed by drowsiness. In April the child was found to have partial paralysis of both legs and of the left arm. On May 3 the patient was brought to Dr. Dandy.

Examination revealed a semistuporous boy who seemed to be in great pain. The pulse varied between 50 and 60. Respiration between 16 and 20. There was bilateral papilledema with hemorrhages and exudate. The swelling of the discs was estimated at 3 diopters. Both sixth nerves were paralysed. There was severe left hemiplegia and paralysis of the right leg.

Two days later the respiratory rate fell to 12 per minute and the pulse rate to 30. The patient was in deep coma. Ventricular puncture was performed and a small amount of fluid escaped under great pressure. The pulse and respiration

improved at once, but within a few hours both respiration and pulse grew slow again and death occurred from failure of respiration.

Post-mortem examination revealed the typical lesions of Schilder's *encephalitis periaxialis diffusa*. The right hemisphere contained a massive lesion and was so greatly swollen that it had herniated under the falx and compressed the left hemisphere. The sulci were obliterated and the ventricles very narrow. There was a well marked pressure cone about the cerebellum. Histological examination of the optic nerve heads and retinae showed that the swelling of the discs was true papilledema and not due to optic neuritis such as is often found in this condition.

Brain Swelling of Unknown Origin: Dandy has described a series of cases in which papilledema and other signs of increased intracranial pressure developed in previously healthy individuals and in which, despite a careful study, the cause remained completely obscure. In these cases no signs of focal damage to the brain can be discovered and the patient's mental condition is usually clear except when the intracranial pressure is very high. The spinal fluid is under increased tension but is otherwise normal. Ventriculography reveals that the ventricles are small and symmetrically placed as if they were compressed by general swelling of the brain. Subtemporal decompression is usually performed and after operation there is, as a rule, herniation through the opening for a few weeks or more. In most cases this herniation disappears and never returns, but in some there may be bulging of the decompression from time to time during a period of years. Except for the possibility of optic atrophy, the prognosis is good.

Similar cases of unexplained cerebral edema have been described by German authors, notably Reichardt who employs the term "*hirnschwellung*." Reichardt makes a sharp distinction between true edema of the brain in which the excess of fluid is found in the interstices of the tissues and *hirnschwellung* in which the fluid is taken up by the cells. It is doubtful whether this distinction can be maintained, however. For our purposes it is sufficient to state that these are cases of swelling of the brain for which no cause can be found even at post-mortem examination.

TYPE DUE TO INCREASED VENOUS PRESSURE

It is well known that obstruction of the venous return from within the cranial cavity is associated with a rise of the cerebrospinal fluid

pressure. Indeed this phenomenon is made use of clinically when Queckenstedt's test is performed. Experimentally it has been found that the cerebrospinal fluid tension is slightly higher than the venous pressure and there is no doubt that the latter is an important factor in determining the rate of formation and absorption of the cerebrospinal fluid. Some patients with failure of the right side of the heart show a marked increase in venous pressure and we may expect that this elevation affects the intracranial tension. Friedfeld and Fishberg investigated 55 cases of failure of the right side of the heart and in all but one there was an elevated cerebrospinal fluid pressure. In one case the manometric reading at lumbar puncture was 450 mm. water and in similar cases Harrison found a patient whose pressure was 520 mm. water. In chronic heart failure the intracranial tension may remain elevated for weeks or months. Venesection or the use of digitalis causes both the venous and cerebrospinal fluid pressures to fall but the latter may remain elevated for several days after a normal venous pressure is obtained. The factors responsible for the rise of cerebrospinal fluid pressure might be the venous engorgment, an increased filtration pressure in the choroid plexuses or actual edema of the brain. Distension of the cervical veins is characteristic of failure of the right side of the heart, and the retinal veins may also be swollen in this condition.

Mediastinal growths or aortic aneurysms may produce great obstruction to the venous return from the head. Ferris has recently studied a group of patients in whom the superior vena cava was compressed. In these cases the intracranial pressure was high and a reading of 490 mm. water was noted in one patient. All had edema of the face, neck and arms but no papilledema was seen. These patients, as well as those with chronic heart failure, had no complaints referable to the state of intracranial tension nor did they have any of the signs usually associated with elevated cerebrospinal fluid pressure. This curious absence of the expected symptoms and signs is difficult to explain. In cases showing edema of the head, neck and upper part of the trunk it might be expected that the brain would also be involved in the edematous process. The extracranial venous system is subject only to the external pressure of the atmosphere whereas a different mechanism obtains within the skull. The increase in venous pressure

will be communicated to the cerebrospinal fluid and as the cranial contents are relatively incompressible the balance of pressures inside and outside the veins may prevent any exudation. The absence of brain edema may be explained in this way but we must note the failure to appear of any signs of intracranial hypertension even though the increase in pressure is marked and long sustained. The increased cerebrospinal fluid pressure in these cases of obstruction to the venous return seems to form an exception to the descriptions which we have given in all other varieties of increased intracranial pressure and the problem needs further investigation.

TYPE DUE TO DISTENTION OF THE VASCULAR CHANNELS

Distention of the venous system within the cranium must play a rôle in the increase of intracranial pressure which occurs in thrombosis of the dural sinuses, but this factor is probably less important than the massive edema of the brain which is the chief cause of the elevation of pressure in such cases. In intracranial arteriovenous fistulae, however, distention of the venous channels seems to be the major element in the elevation of pressure which occurs in a certain percentage of cases. Cushing and Bailey make the following statement in this connection:

"However, the mechanism of the condition (increased intracranial pressure in arteriovenous fistulae) must be somewhat different from that which produces choked disc in the ordinary space-constricting brain tumor. For here is a lesion which takes up room, not because of its size as a foreign body, not because it causes cerebrospinal fluid stasis, but because it actually increases the amount of blood in the intracranial chamber by pumping arterial blood directly into the cerebral veins. . ."

When one thinks of the enormous dilatation of the cortical and meningeal veins which sometimes occurs in such cases, it is evident that this explanation is an adequate one. It must also be remembered that the pressure within the veins is increased for they are receiving blood under arterial pressure. Cushing and Bailey found evidences of increased intracranial pressure in thirteen out of fourteen cases, but in only three cases was there pronounced elevation of pressure. In Dandy's series of eight cases only two showed definite papilledema.

As a rule the intracranial pressure is only moderately increased and tends to remain at approximately the same level for long periods. The following case is representative in this regard:

Case History (U-65744). Focal convulsions beginning in right hand at the age of 7 years. At age of 14 years headache and at 16 years papilledema. Loud intracranial bruit. Operation revealed arteriovenous fistula between left middle cerebral artery and Rolandic vein. Subtemporal decompression through which brain herniated during next few years.

L. U., was regarded as a normal child until the age of 7 years when she began to have convulsive seizures beginning in the right hand. These continued at variable intervals. At the age of 14 years the child began to complain of headaches and of throbbing in her head.

Examination revealed engorgement of the retinal veins but no papilledema. There was a loud systolic bruit heard best over the left side of the cranial vault. There was no weakness of the right hand but mild sensory changes were found which suggested cortical anesthesia. Otherwise the neurological examination was negative. Roentgenographic examination of the skull disclosed a large groove in the left parietal bone, corresponding to the position of the Rolandic vein.

During the next two years the patient continued to have frequent seizures. The headaches grew more frequent and more severe. She vomited occasionally. At the age of 16 years the patient was reëxamined and found to have bilateral papilledema. Dr. Dandy explored the left hemisphere and found great dilatation of the left Rolandic vein which pulsated with each heart beat and which was filled with bright red blood. Large tortuous veins extended both anteriorly and posteriorly from the Rolandic vein. It was decided that there was a fistula between the middle cerebral artery and the Rolandic vein. No attempt was made to extirpate the lesion, but decompression was performed. More than 3 years have elapsed since the operation and throughout this period there has been herniation of the brain through the defect in the bone. The defect always bulges but is not very tense. The headaches have ceased, but the convulsive seizures persist. The bruit is still audible.

TYPE DUE TO DIMINISHED CAPACITY OF THE CRANIUM

In oxycephaly the increased intracranial pressure is due to a defect in the cranium. The membranous bones of the vault fail to grow normally, the coronal and sagittal sutures close early and the capacity of the cranium is reduced. During early childhood the rapid growth of the brain causes definite elevation of intracranial pressure which gives rise to increased convolutional markings in the bones of the vault, to bilateral exophthalmos and usually to papilledema which is followed by optic atrophy. The ventricles are small and compressed.

Measurements of spinal fluid pressure may give readings of over 400 millimeters of water. If decompression or some similar operation is carried out early enough, optic atrophy may be prevented. It is of interest that papilledema and optic atrophy occur only early in childhood, usually before the age of five years. If the child has good vision at the age of ten years, no fears of optic atrophy need be entertained. Evidently the increased intracranial pressure exists only during the period of active growth of the brain. We have found normal spinal fluid pressures in two adults who exhibited the usual sign of oxycephaly.

Case History. Oxycephaly with exophthalmos, headaches and papilledema during childhood. Later, headaches ceased. At age of 34 years, optic atrophy and convolutional atrophy of skull. Spinal fluid pressure normal.

W. W. was born with a deformed head of oxycephalic shape. During early childhood he was subject to severe headaches and examination during this period is said to have shown mild papilledema. After adolescence the headaches ceased.

At the age of 34 years examination disclosed an oxycephalic head sloping up to an apex at the vertex and an extreme degree of bilateral exophthalmos. There was some secondary optic atrophy and vision was reduced to about 20/50 on either side. Roentgenograms of the skull showed pronounced convolutional markings in the cranial vault and obliteration of the coronal and sagittal sutures. The spinal fluid pressure was 140 millimeters of water.

SUMMARY AND CONCLUSIONS

1. Increased intracranial pressure may be estimated by manometric readings of the spinal or intraventricular pressure but such studies involve a certain element of danger and are sometimes misleading. Moreover, there is reason to believe that in states of increased intracranial pressure there are spontaneous fluctuations in pressure of such a magnitude that one reading is no more valuable than one determination of the blood pressure in a case of arterial hypertension. The clinical signs and symptoms give more reliable information about the *average* level of intracranial pressure.

2. Increased intracranial pressure may be due to a large number of diseases but is commonly produced by only a few mechanisms. Six types are described below:

3. Hydrocephalus is the commonest cause of increased intracranial pressure. It is almost invariably due to obstruction of the channels

through which the cerebrospinal fluid flows. When obstruction occurs suddenly there is at once great elevation of intracranial pressure which may prove fatal within a short time. In other cases there is a gradual compensation and the pressure may fall to a normal or nearly normal level. This compensation, we believe, is due to reduction of secretory activity of the choroid plexus resulting from compression by the high pressure in the ventricles. In some cases in which the obstruction develops slowly it is apparently possible for compensation to occur at the same pace so there is never any great elevation of pressure. Compensated hydrocephalus is seen most frequently in cases of congenital origin. At any time compensation may fail and sudden elevation of pressure may reappear. This is often induced by ventricular puncture which seems to cause activation of the choroid plexus, but may also occur spontaneously and for no apparent reason.

4. Masses within the cranium may cause elevation of intracranial pressure without hydrocephalus. A slowly growing tumor may attain a very large size, however, without causing definite signs of increased intracranial pressure. The brain is very soft and plastic and when the patient is in the upright position the normal pressure in the veins and in the ventricular system is very low. The growth of the tumor is compensated by moulding of the brain and by reduction of the fluid content of the ventricles, and of the subarachnoid and perivascular spaces, and by diminution of blood in the veins. It is not until the capacity for compensation is exhausted that definite elevation of intracranial pressure results though there are signs of compression of the brain long before. The common subdural hematoma is taken as an example of this type of increased intracranial pressure.

5. A third cause of increased intracranial pressure is edema or swelling of the brain. This may be due to a number of causes including trauma, venous obstruction, and intoxications and is associated with certain hypertensive states such as acute glomerular nephritis, lead encephalopathy and eclampsia. Both local and general edemas are found in this group. In the former some compensation is possible, for the normal parts of the brain may yield to the compression and prevent a general increase of pressure within the cranium until the edema becomes extensive.

6. In certain chronic arterial hypertensions of adults, especially

in "malignant" types, there may be elevation of intracranial pressure. It is doubtful whether this is a result of edema of the brain as it is in acute glomerular nephritis. The relation of the increased intracranial pressure to the hypertension is obscure, for the intracranial pressure does not parallel the systolic arterial pressure although it is claimed that it has a tendency to vary with the mean arterial pressure.

7. Obstruction of the venous drainage of the head in failure of the right side of the heart and in certain cases of mediastinal tumor is another cause of increased intracranial pressure. In some cases the spinal fluid pressure may excede 500 mm. of water and remain at a high level for long periods. Nevertheless, it is claimed that the patients have no papilledema and complain of none of the usual symptoms of increased intracranial pressure.

8. Increased intracranial pressure is found in some cases of arteriovenous fistula within the cranium. In this case the distention of the blood vessels seems to be responsible. The pressure may remain at approximately the same level for a number of years.

9. A rare cause of increased intracranial pressure is diminished capacity of the cranium. This is found most frequently in oxycephaly. The normal growth of the brain is resisted by the rigid walls of the small cranium and elevation of intracranial pressure results. The pressures seems to persist only during the first five years of life when the growth of the brain is most vigorous.

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MORPHOLOGIC CHANGES IN THE BLOOD OF PIGS ASSOCIATED WITH DEFICIENCY OF WATER-SOLUBLE VITAMINS AND OTHER SUBSTANCES CONTAINED IN YEAST

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With the purpose of observing the changes in the blood and the nervous system of young animals resulting from deficiency of water-soluble vitamins and other substances contained in yeast, young pigs have been raised since early life on an artificial diet consisting of casein, sucrose, lard, cod liver oil, a mineral mixture, ascorbic acid and yeast. As these animals became accustomed to the diet, the quantity of yeast was gradually reduced and in its place the synthetic substances thiamin chloride (B_1), riboflavin and nicotinic acid were given, either separately or in various combinations. These investigations were undertaken in an attempt to reproduce in animals a condition similar to pernicious anemia in man. One of us has recently described the hematopoietic effect of yeast in cases of this disease (1). A preliminary report regarding the occurrence of sensory neuron degeneration in these animals has already been published (2). It is the object of this report to describe the anemia which was observed. It was characterized by the presence of macrocytes and many immature red corpuscles as well as by hyperplasia of the bone marrow, and could be relieved by the administration of yeast.

REVIEW OF LITERATURE

McGowan and Sinclair (3) found that young pigs kept on a ration of corn, fish meal and draff became ill with anemia, jaundice and liver damage. The anemia was described as macrocytic and the femoral

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marrow was red and cellular. Reticulocyte increases occurred following administration of raw liver, the anemia disappeared and the bone marrow became normal. Miller and Rhoads (4) fed swine weighing 11 to 13 Kg. a modified canine-blacktongue-producing diet. A symptom complex developed, characterized by oral mucous membrane lesions, diarrhea, achlorhydria, motor weakness and anemia. In 9 animals the anemia was considered to be macrocytic whereas in 6 others it was called microcytic. Reticulocytosis followed the injection of liver extract, whereas administration of iron did not prevent the development of the microcytic anemia. The gastric juice and the livers of the animals which developed macrocytic anemia were found to be deficient in substances effective in the treatment of pernicious anemia.

Birch, Chick and Martin (5) gave pigs, 81 to 84 days of age, a maize diet not supplemented by yeast. A mild, progressively increasing anemia developed. Chick and her co-workers (6) have just reported in more detail on the effects of such a diet when supplemented by aneurin (B_1), lactoflavin, and nicotinic acid as well as eluate and filtrate fractions of liver. When the diet was supplemented by all of these substances or by 4 per cent brewers' yeast, no anemia was observed after 10 weeks. The effect of deprivation of filtrate fraction was to produce a moderate anemia of the normocytic type. One pig subsequently given filtrate fraction failed to show improvement in 3 weeks. The effect of deprivation of the eluate fraction was to produce well marked microcytic, hypochromic anemia which could be corrected in 7 weeks by administration of this fraction.

Attempts have been made to produce a "conditioned deficiency" in pigs similar to that occurring in pernicious anemia, by means of gastrectomy. Bence (7), Maison and Ivy (8) and Geiger, Goodman and Claiborn (9) found that microcytic, hypochromic anemia developed. The last named investigators observed macrocytic anemia in one sow which had been operated upon 30 months before. Bence stated that this occurred in his animals in the second or third year following gastrectomy but his data are not convincing. The bone marrow in his animals was described as resembling that of pernicious anemia. The livers of the agastric animals studied by Geiger, Goodman and Claiborn, gradually lost their reticulocytogenic power but

this loss was not complete until 18 months following gastrectomy. Young pigs 6 weeks old, gastrectomized by Petri, Norgaard and Bing (10), not only developed hypochromic microcytic anemia, but growth was arrested and extensive changes occurred in the skin and nervous system.

Certain pertinent observations in other animals may be mentioned. In rats, diets low in the vitamin B complex have led to the development of simple hypoplastic anemia with leukopenia (11) or hypochromic anemia (12). Anemia in such animals could be prevented by the addition of yeast to the diet and it was cured by giving lean beef muscle or liver but not by giving iron, copper or vitamins A and D (13). Anemia occurred irregularly in dogs fed a diet low in protein, high in fat and carbohydrate and containing only the vitamin B present in rice polishings (14); anemia also developed in dogs in which chronic recurrent blacktongue was produced (15, 16). The anemia was irregularly macrocytic, could be relieved by meat, yeast or liver fed by mouth but did not respond to liver extract given parenterally. Margolis and others (17) have recently stated that macrocytic anemia failed to develop in dogs given a blacktongue-producing diet which was supplemented by thiamin, riboflavin and nicotinic acid. When a "sprue diet" such as that used by Miller and Rhoads was fed gastrectomized dogs, hypochromic microcytic anemia developed (18).

Fouts and others (19) recently reported that, when given a diet in which the rat anti-dermatitis factor (B_6) was the only missing component, puppies developed severe, microcytic hypochromic anemia which could be relieved by the addition of this factor to the diet. György, Robscheit-Robbins and Whipple (20) have presented evidence that the administration of riboflavin causes a definite increase in hemoglobin production above the basal level in standardized anemic dogs. According to György (21) nicotinic acid is also concerned in hematopoiesis, for the administration of this substance prevented the development of panmyelophthisis previously observed by him in rats (22).

In monkeys, Wills and Stewart (23) produced macrocytic anemia by the use of a diet low in protein and in vitamin B complex. The anemia could be cured by the administration of autolysed yeast and to some extent by the use of a filtrate fraction of yeast (24). Johnstone

and Reed (25) observed progressive anemia of an undetermined nature, and leukopenia in two monkeys given a diet deficient in the B complex. Langston and others (26) found that a diet deficient in B factors other than thiamin, riboflavin and nicotinic acid, caused normocytic anemia and leukopenia in monkeys and this could be prevented by the administration of yeast or liver extract.

That a diet deficient in that part of the B complex other than B_1, riboflavin and antidermatitis concentrate, leads to the development of anemia even in pigeons, is indicated by the recent report of Hogan and others (27).

As in swine, gastrectomy in other animals has been followed by microcytic anemia (28, 29, 30, 31) and this type of anemia has also followed extirpation of the duodenum (32) and even removal of the duodenum plus a portion of the jejunum in addition to the stomach (33, 34). Even when the livers of gastrectomized dogs were injured by means of carbon tetrachloride the anemia was macrocytic only in the terminal stages (35).

EXPERIMENTAL METHODS

The pigs were farrowed at the experimental farm of the United States Department of Agriculture, Beltsville, Maryland. This report is concerned with 21 pigs derived from 4 litters as follows: litter VII, Duroc-Jersey, pigs 4-60, 4-63, 4-64; VIII, Poland China, pigs 4-78, 4-79, 4-80, 4-82; IX, crossbred, Duroc-Jersey and Landrace, pigs 4-85 to 4-90, inclusive; X, Poland China, pigs 4-91 to 4-98, inclusive. These animals were received in Baltimore at 15 to 17 days of age, except 4-60 which was 10 days old, and 4-63 and 4-64 which were 23 days of age. They were housed out of doors in pens consisting of a shelter with a wood floor and a runway floored by tile. Two or three pigs on the same type of diet were housed together in each pen. They were fed three times daily at first and less often as they became older, until only one feeding was given daily. The basal diet consisted of casein, sucrose, lard, cod liver oil, salt mixture and ascorbic acid as described elsewhere (1, 36). All the pigs except 4-90 and 4-95 were given an amount furnishing 156 cal. per Kg. daily. These two pigs served as "inanition controls" and were given only as much food as the pigs with the poorest appetites had consumed the day before. This resulted in their being given from 52 to 115 cal. per Kg. (average about 70 cal.) from the age of 105 days.

This diet was considered adequate except for its content of water-soluble B vitamins. At first the diet of all the pigs was supplemented with dry brewers' yeast (Northwestern Yeast Co., Chicago), 3 gm. per Kg. daily. This or a greater amount was given throughout the period of observation to 6 animals while in the

remainder the amount of yeast was gradually reduced to amounts even as low as 0.1 gm. per Kg. In four animals thiamin chloride was given as the yeast was withdrawn; in five, thiamin and riboflavin were substituted; in three, thiamin and nicotinic acid; and in three, thiamin, nicotinic acid and riboflavin were used.

The supplements were given as follows: yeast was mixed with the food daily. The synthetic vitamins were given twice a week in solution by intramuscular injection (thiamin, riboflavin) or were dropped by means of a pipet into the back of the mouth of each pig (nicotinic acid, riboflavin). These substances were made up in solution freshly each time. Except in three pigs (4-60, 4-63, 4-64), the amounts given were somewhat greater than the quantities of these substances contained in the yeast given the control animals. Thiamin chloride (Mead Johnson and Co., Winthrop Chemical Co.) was given in amounts of 1 mg. per Kg. body weight twice a week. Half this amount of riboflavin (Merck and Co.) was used. The quantity of nicotinic acid (S. M. A. Corporation) given was 7 mg. per Kg. twice a week.

As far as possible pigs of each experimental group were derived from more than one litter.

Blood was obtained by venepuncture, the brachial veins usually being used. This was collected in a mixture of ammonium and potassium oxalate (6.5 mg. of the former and 3.5 mg. of the latter per 5 cc. blood) which caused no change in the size of the red corpuscles. Venous blood was used for all quantitative measurements. Hemoglobin was measured by means of a Newcomer hemoglobinometer restandardized by the oxygen capacity method. Volume of packed red cells was measured by means of a hematocrit and the mean volume and hemoglobin content of the red corpuscles were calculated as described elsewhere (37). Icterus index was estimated, and leukocyte, platelet and reticulocyte counts were made on the venous blood. Blood smears were made from blood drawn from the ear. When daily reticulocyte counts were made the blood was also obtained from the ear. Erythrocyte diameter measurements were made by means of an ocular micrometer.

Gastric analyses were made from time to time. The usual procedure was to inject histamine (.01 mg. per Kg.) in the morning before the pigs had been fed, and to withdraw the gastric contents by means of a stomach tube a half hour later. More recently the fasting contents have been removed by stomach tube before histamine was injected. The gastric juice was titrated with $\frac{N}{20}$ NaOH using Töpfer's reagent and phenolphthalein as indicators.

Complete autopsies were made of each animal. In a few instances bone marrow was obtained during life by sternal puncture. At autopsy specimens of the sternum, ribs, vertebrae, femora and tibiae were obtained. The following procedure was finally found to be the most satisfactory for obtaining specimens of bone marrow and was carried out in about half the pigs: the femur was cut longitudinally and the soft marrow which is found in the shaft was cut out so that a large portion of marrow free of bone was obtained. Since this did not require

decalcification, excellent sections from the midshaft, and the proximal and distal portions of the shaft were obtained.

RESULTS

This report will be confined to a description of the findings in the blood, bone marrow and gastric contents. Details concerning the growth of the animals will be published elsewhere (36), and an account of the pathologic changes in the nervous system of the animals will be presented when the histologic examinations have been completed. It may be stated here that when the basal diet was supplemented with yeast, growth was good. When yeast was withdrawn growth became slower and even when nicotinic acid as well as riboflavin and thiamin chloride were given, ataxia, such as has already been described (2), developed and growth eventually became impaired. It was evident that these synthetic substances did not make up for all the growth-promoting and nerve-protecting substances present in yeast.

The results of the blood examinations are summarized in Table I and in figure 1. In figure 1 only the values for volume of packed red cells have been plotted because these are less subject to technical error than the red cell count or hemoglobin.

A. Basal diet supplemented by 3 gm. or more of yeast per Kg. body weight daily

In this group of pigs, which consisted of representatives of each of the litters used, growth was good and, in spite of fluctuations (figure 1), the blood of these animals was essentially unchanged after 130 to 223 days on this diet. Not only did anemia fail to develop, but the mean volume and diameter of the red corpuscles remained unchanged and the blood smears showed no increase in the degree of anisocytosis or poikilocytosis. Like other observers (38, 39) we have noted in the blood of apparently normal young pigs more anisocytosis than is seen in the blood of normal adults, as well as occasional polychromatophilia. Bizarre forms of poikilocytes may be produced unless the blood smears are well made. Howell-Jolly bodies may be seen, in the absence of anemia, as late as 6 months of age. We have observed occasional normoblasts in the blood smear as late as 4½ months of age and polychromatophilia as late as 8 months. Reticulocytes in

TABLE I

Blood of Pigs Given Basal Diet Plus Various Supplements

A. Brewers' yeast, 3 gm. or more per Kg. body weight daily

Pig No. & Sex	4-78 ♂		4-89 ♀		4-90 ♀		4-91 ♂		4-95 ♀		4-96 ♀	
Age (days)	51*	213	40	170	40	263	41	216	41	208	48	216
Weight (Kg.)	10.0	104.0	4.9	78.0	4.2	51.7	3.9	63.5	4.6	42.0	4.6	61.0
R.B.C. (millions)	9.38	8.82	8.30	8.05	7.50	8.15	7.07	7.85	7.56	7.35	8.60	7.34
Hgb. (gm.)	13.7	14.1	15.5	16.2	14.1	15.1	13.5	16.2	14.3	13.5	13.7	14.9
Vol. pkd. R.B.C. (cc.)	43.2	41.5	51.0	51.5	45.0	47.0	43.5	48.5	44.8	43.0	46.2	45.5
M.C.V. (c.μ.)	47	47	61	64	60	58	62	62	59	59	54	62
M.C.D. (μ.)	5.47	5.44	5.74	5.93	5.42	5.54	5.66	5.45	5.61	5.40	5.69	5.23

B. Thiamin chloride (B_1), 1.0 mg. per Kg. twice a week, gradually reduced amounts of yeast

Pig No. & Sex	4-60 ♂		4-87 ♀		4-88 ♀		4-94 ♀					
Age (days)	56	234	58	156	61	114	55	102				
Weight (Kg.)	4.8	31.0	7.0	17.2	9.6	18.1	6.3	9.0				
R.B.C. (millions)	6.76	6.15	8.40	6.70	7.27	6.75	7.48	3.89				
Hgb. (gm.)	11.2	12.8	17.0	12.0	14.9	12.0	13.7	7.1				
Vol. pkd. R.B.C. (cc.)	35.6	36.0	50.5	37.0	45.1	36.5	43.0	22.0				
M.C.V. (c.μ.)	52	58	60	56	62	54	58	57				
M.C.D. (μ.)	5.49	5.34	5.78	5.57	5.68	5.64	5.51	5.64				

C. Riboflavin, 0.5 mg., and thiamin chloride, 1.0 mg. per Kg. twice a week, reduced amounts of yeast

Pig No. & Sex	4-63 ♀		4-64 ♀		4-85 ♀		4-97 ♀		4-98 ♀			
Age (days)	56	205	56	231	60	136	55	132	55	103		
Weight (Kg.)	7.8	18.5	7.6	26.0	7.7	25.9	6.2	11.8	6.0	12.0		
R.B.C. (millions)	8.00	7.52	6.79	7.43	7.59	6.41	6.62	5.05	7.68	7.15		
Hgb. (gm.)	14.5	12.4	13.1	10.4	14.1	14.1	13.1	8.7	13.1	11.5		
Vol. pkd. R.B.C. (cc.)	43.5	39.2	37.0	36.0	43.0	40.0	37.3	33.2	41.0	35.3		
M.C.V. (c.μ.)	54	52	54	48	52	62	56	66	53	50		
M.C.D. (μ.)	5.70	5.52	5.62	5.65	5.64	6.05	5.74	6.15	5.44	5.25		

D. Nicotinic acid, 7 mg. and thiamin chloride, 1.0 mg. per Kg. twice a week, reduced amounts of yeast

Pig No. & Sex	4-86 ♀		4-92 ♂		4-93 ♂							
Age (days)	60	163	54	124	54	124						
Weight (Kg.)	8.7	34.0	6.4	14.0	6.5	14.0						
R.B.C. (millions)	7.70	4.97	8.15	6.03	7.29	6.08						
Hgb. (gm.)	13.1	9.9	13.1	11.5	13.1	12.8						
Vol. pkd. R.B.C. (cc.)	46.0	30.0	41.1	33.8	41.5	36.8						
M.C.V. (c.μ.)	60	60	50	56	57	59						
M.C.D. (μ.)	5.85	6.15	5.31	5.38	5.77	5.69						

E. Nicotinic acid, 7 mg., thiamin chloride, 1 mg., riboflavin, 0.5 mg. per Kg. twice a week, reduced amounts of yeast

Pig. No. & Sex	4-79 ♀		4-80 ♀		4-82 ♀							
Age (days)	51	213	51	134	52	163						
Weight (Kg.)	11.1	47.6	9.2	22.0	10.0	32.2						
R.B.C. (millions)	9.88	6.43	9.96	6.22	9.69	5.67						
Hgb. (gm.)	15.9	11.5	16.8	10.3	17.2	11.0						
Vol. pkd. R.B.C. (cc.)	48.2	34.5	53.3	30.5	53.0	34.0						
M.C.V. (c.μ.)	48	54	54	50	55	60						
M.C.D. (μ.)	5.10	5.55	5.70	5.54	5.44	5.67						

* For each pig are given the blood values found (1) early in the experiment (Group A) or at the time when the quantity of yeast was first reduced below 3 gm. per Kg. (Groups B to E) and (2) at a time near death when the quantity of yeast supplied was very low. This did not always correspond with the time of death because some pigs were given large amounts of yeast before they died.

R.B.C. refers to red cell count in millions per c.mm.; Hgb. refers to hemoglobin in gm. per 100 cc. blood; vol. pkd. R.B.C. refers to volume of packed red cells in cc. per 100 cc. blood; M.C.V. indicates mean corpuscular volume, in cubic microns; M.C.D. indicates mean corpuscular diameter, in microns.

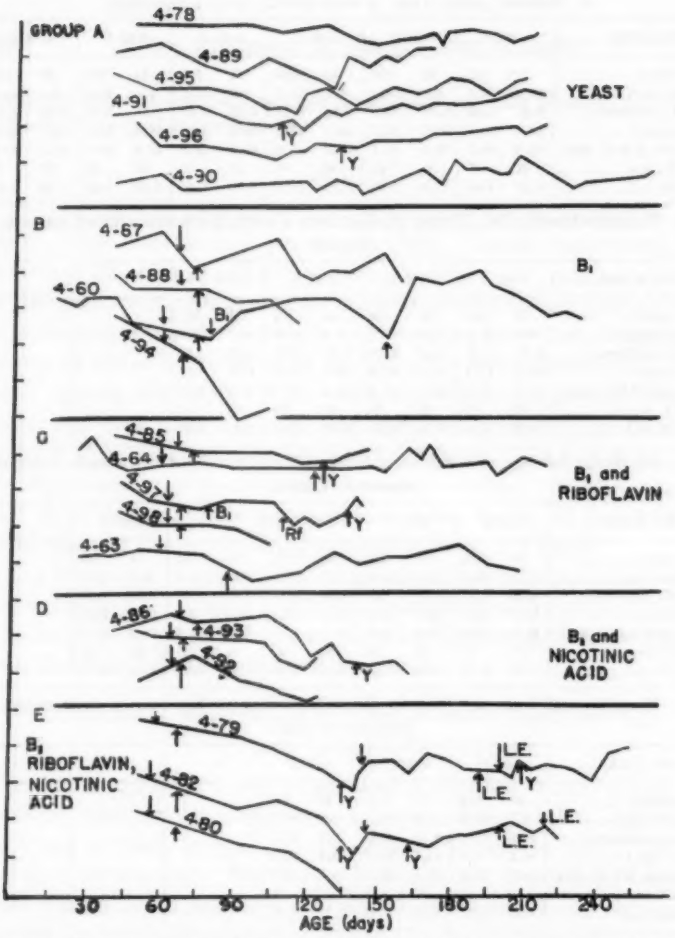

FIG. 1. Volume of packed red cells in the blood of pigs given an artificial diet, (A) supplemented by 3 or more grams brewers' yeast per Kg. body weight daily; or (B to E) provided with reduced amounts of yeast, but supplemented by (B) thiamin chloride, (C) thiamin and riboflavin, (D) thiamin and nicotinic acid, and (E) thiamin, riboflavin and nicotinic acid.

Each division on the ordinate represents 10 cc. of packed red cells per 100 cc. blood. Arrows pointing downwards indicate the time when the quantity of yeast was reduced; arrows pointing upwards indicate when administration of synthetic vitamins was commenced. Exceptions are marked: Y (yeast), B_1 (thiamin), Rf (riboflavin) or L.E. (liver extract). In all instances an arrow pointing upwards indicates commencement or increase, an arrow pointing downwards refers to decrease or discontinuation.

the control group of pigs fluctuated irregularly between 0.4 and 2.5 and even 3.5 per cent; occasionally values as high as 5.5 per cent were found. This tendency for immature forms of the red cell series to appear in the blood of normal pigs even as late as 8 months of age, must be kept in mind when anemia is being studied in young animals of this species.

Anemia also failed to develop in the animals (4-90, 4-95) given reduced amounts of food supplemented by 3 grams of yeast ("inanition controls").

Two pigs of the control series were given 6 instead of 3 grams of yeast per Kg. daily from the ages of 108 and 133 days, respectively. In both instances (4-91, 4-96), their blood counts rose gradually to levels 18 and 14 per cent, respectively, higher than they had been (figure 1).

All of these pigs were killed. The vertebral, sternal and rib marrow was moderately cellular. The proximal and distal portions of the femur were uniformly red and made of cancellous bone. The middle two-thirds of the femur was soft and completely devoid of bone. Most of this soft marrow was fatty. Only a few small red patches were scattered here and there. At the edges of the soft marrow, in proximity to the bone a reddish zone of varying thickness (2–5 mm.) was present. In these areas of the soft marrow active blood formation was observed on microscopic examination. The remainder of the soft marrow contained very few cells. (Plate I, figure 3.)

B. Basal diet with reduced quantities of yeast, supplemented by B_1

The pigs in this group were derived from 3 litters. At the ages indicated by the arrows pointing downwards in figure 1B, at a time when the pigs seemed to have been successfully weaned on the artificial diet, the quantity of yeast given the pigs was gradually reduced from 3 gm. per Kg. body weight daily to levels as low as 0.5 to 0.1 gm. In all four pigs anemia developed, although its degree differed considerably. Pig 4-60 was already somewhat anemic when the quantity of yeast was reduced. The anemia became more severe. Two injections of thiamin chloride of 25 mg. each, were followed by a rapid disappearance of anemia (see figure 1) as well as improvement in the rate of growth. Two and a half months later, with continued

reduction in the amount of yeast, anemia was once more well marked (R. B. C. 5.05, vol. packed red cells 31.0 cc.). Regular injections of thiamin were commenced at this time, 0.25 mg. per Kg. twice each week. As had happened before, growth improved and the anemia disappeared. On this occasion the improvement was even more striking than before. In the end, however, in spite of continued administration of thiamin, growth ceased, loss of weight occurred, anemia developed again and the animal died. A detailed account of the changes in the nervous system in this animal has been published elsewhere (2). Signs of ataxia commenced when the blood was normal (182 days of age).

The mean corpuscular volume, at the three points of most severe anemia, was 57, 62 and 58 c.μ., respectively. These values are not significantly higher than the values found in this pig when the experiment was commenced.

In the remaining 3 pigs of this group, the regular biweekly administration of thiamin chloride (1 mg. per Kg. twice a week) was commenced a few days after the quantity of yeast was first reduced. Anemia developed in all, although in 4-87 a temporary increase occurred in the red cell count, hemoglobin and volume of packed red cells following administration of thiamin, like that observed in 4-60 (figure 1). Unlike 4-60, however, in this pig growth had not yet commenced to slow up as the result of withdrawal of yeast. Growth in 4-88 was good until 3 weeks before death. In 4-94, on the other hand, growth had never been good. Very severe anemia developed in this animal. At the age of 91 days, when the yeast had been reduced to 0.5 gm. per Kg. and the erythrocyte count was 3.13 millions, the M. C. V. was 56 c.μ., and polychromatophilia and macrocytosis were observed in the blood smear. Five days later, the macrocytosis was even more striking, there was marked polychromatophilia and occasional stippling and Howell-Jolly bodies, as well as 14 normoblasts and macroblasts per 100 leukocytes were observed. Reticulocytes numbered 6.6 per cent and increased as high as 13.2 per cent. A moderate number of microcytes were also present so that no increase in the mean volume or mean diameter of the red cells was observed.

All these pigs died, 4-60 presumably of malnutrition; 4-87 of a gastrointestinal infection with beginning peritonitis; 4-88 of lobular pneumonia with multiple abscesses in the lungs; 4-94 of lobar pneu-

monia. The bone marrow of 4-60 and 4-88 was much like that of the control animals; that of 4-87 showed slight hyperplasia; the femoral marrow of 4-94 was gelatinous and maroon colored and definitely hyperplastic. The spleen of this pig showed numerous foci of extramedullary blood formation. (Plate I, figure 7.)

C. Basal diet with reduced amounts of yeast, supplemented by B_1 and riboflavin

In pigs 4-63 and 4-64, reduction of the quantity of yeast was begun some time before supplements of the synthetic vitamins were added. In both, moderate anemia appeared before the growth rate had been greatly affected. Examination of the blood smears revealed a moderate number of macrocytes, some of them quite large. Administration of thiamin chloride (0.5 mg. per Kg. twice a week) and riboflavin (0.2 mg. per Kg. twice a week) was followed by a gradual decrease of anemia, although the rate of growth was not restored to normal. When these animals finally died, the erythrocyte values were little below those found when reduction of yeast was begun. Yet in the blood smear of 4-64 a moderate number of macrocytes and several Howell-Jolly bodies as well as giant platelets were observed. A sternal puncture in this animal 5 weeks before death revealed more immature erythrocytes, some quite large, than had been observed in the bone marrow of control animals. At autopsy some hyperplasia of the marrow was found. The bone marrow of 4-63 was much like that of the control pigs. Broncho-pneumonia was found in this pig but no evidence of infection was seen in the autopsy of 4-64.

In pigs 4-85, 4-97 and 4-98, thiamin and riboflavin were given in larger doses (1.0 and 0.5 mg. per Kg. twice a week), and this was done as soon as reduction in the quantity of yeast was commenced. In the first of these animals growth was good until near the time of death. Very little anemia developed. Growth was poor in pigs 4-97 and 4-98. In these animals, before the quantity of yeast had been reduced below 3 gm. per Kg., some anemia had already developed, but as the yeast was reduced and was replaced by thiamin and riboflavin, there was little further change in the blood in either animal for 38 and 20 days, respectively. The condition of these two pigs at this stage was the same, both having ceased to grow after 90 to 100 days of age. They were thin and poorly nourished as well as ataxic. The

life of 4-97 was prolonged, however, probably as the result of the use of great amounts of thiamin and riboflavin. Thiamin was given in amounts of 2.5 mg. per Kg. daily. This failed to relieve the anemia, and the growth rate was not improved. At the age of 109 days, the red cell count was 5.80 millions, the volume of packed red cells 31.3 cc. The quantity of riboflavin was then increased to 5 mg. per Kg. daily and it was given by stomach tube instead of by intramuscular injection. Anemia became even more marked (R. B. C. 5.25, volume packed red cells 29.5 cc.) and numerous macrocytes, some polychromatophilic and very large, appeared in the blood. The mean corpuscular volume increased to 66 c.μ., the erythrocyte count dropped to 5.0 millions. Giant platelets appeared in the blood smear. The quantity of yeast was increased to 3 gm. per Kg. during the last few days of this pig's life. Reticulocytes increased as high as 18.5 per cent and the anemia became less marked but the pig died nevertheless.

Death resulted from bronchopneumonia in each of these 3 pigs. In 4-97 several abscesses were also present in the legs. The femoral marrow of each was red and gelatinous. Hyperplasia was more marked in 4-98 than in 4-85 and it was very marked in 4-97.

The microscopic appearance of the femoral marrow of 4-97 (Plate I, figures 4 and 5) was typical of that of the marrow of other animals in which hyperplasia was observed, although the degree of hyperplasia varied in the different animals as already noted. Numerous immature red and white cells replaced the fat. A number of clearly recognizable mature forms of the leucocytic series (neutrophilic juvenile leukocytes, eosinophils) were present but in addition many immature forms were observed. Instead of the preponderance of leukocytes as compared with nucleated red cells found in the marrow of the control animals, there was a distinct increase in the proportion of cells of the erythrocyte series. Some of the latter resembled "megaloblasts" although we have found it difficult to be certain of this. A similar preponderance of immature cells was observed in smears made of this marrow. Magalokaryocytes were present in normal numbers.

D. Basal diet with reduced amounts of yeast, supplemented by B_1 and nicotinic acid

In the 3 pigs of this group, growth was much better than in groups B or C. The changes in the blood were also more uniform. As the

quantity of yeast was reduced, anemia developed, in spite of the administration of thiamin and nicotinic acid. In 4-86 the mean corpuscular volume increased to as high as 68 c.μ., the mean diameter to 6.34 μ. and many macrocytes as well as polychromatophilia and Howell-Jolly bodies (0.6 per cent) appeared in the blood smear. The quantity of yeast was increased to 3 gm. and later to 5.4 gm. per Kg. The reticulocytes increased to 9.4 per cent on the eleventh day following the increase of yeast, the mean corpuscular volume decreased to 60 c.μ. and the blood smear became more normal in appearance. This change occurred even though a significant increase in the erythrocyte count failed to occur. Death resulted from cardiac failure. In 4-92 an increase in mean corpuscular volume did not occur but macrocytes appeared in the blood smear, occasional normoblasts were found and large numbers of Howell-Jolly bodies appeared. The changes in the blood of 4-93 were similar but somewhat less marked. In both animals death resulted from lobular pneumonia. Moderate hyperplasia was found in the bone marrow of all three pigs.

E. Basal diet with reduced amounts of yeast, supplemented by B_1, riboflavin and nicotinic acid

In the 3 pigs of this group, growth was good throughout most of their lives. Severe anemia developed in each animal as the quantity of yeast was reduced, in spite of the administration of the 3 synthetic vitamins, thiamin, riboflavin and nicotinic acid. The administration of yeast was followed by a decrease in the degree of anemia.

In 4-79 the red cell count decreased to 4.97 millions and the volume of packed red cells to 29.0 cc. after 84 days on reduced quantities of yeast. The mean corpuscular volume increased from 48 to 59 c.μ. (figure 2), the mean cell diameter increased from 5.10 to 5.69 μ., the blood smear showed many macrocytes, marked poikilocytosis and polychromatophilia, a moderate number of Howell-Jolly bodies (1.7 per cent) and 26 nucleated red corpuscles per 100 leukocytes (figure 2, Plate I). A number of giant platelets were observed in the smear. The pig's appetite was poor and there was diarrhea as well as slight ataxia. The quantity of yeast was increased to 3 gm. per Kg. The reticulocytes were found to be 28.4 per cent on the seventh day thereafter. Unfortunately these had not been enumerated in the preceding days. Their number decreased to 13.4 per cent on the

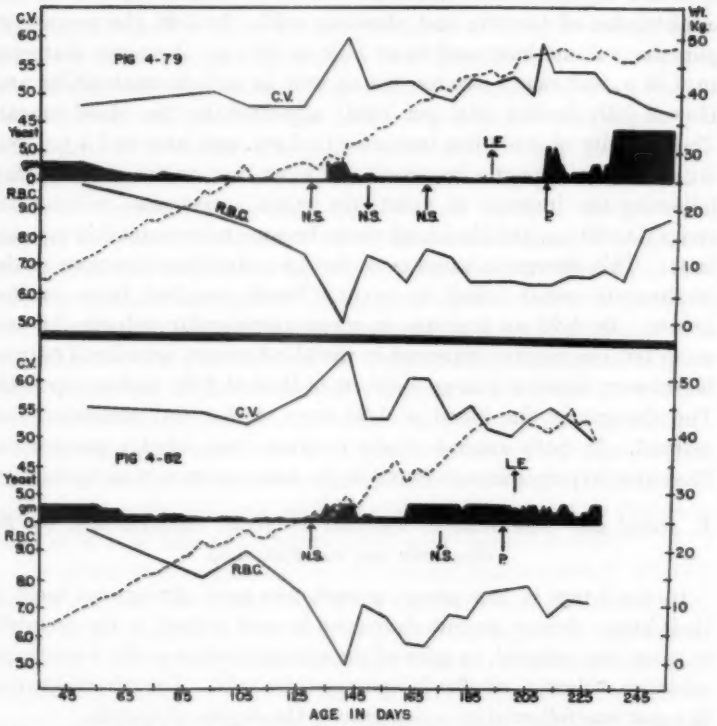

FIG. 2. Development of anemia in two pigs given the artificial diet supplemented by varied amounts of yeast and regular quantities of thiamin chloride, riboflavin and nicotinic acid. As the amount of yeast was reduced anemia and macrocytosis, as well as ataxia developed. As the quantity was increased, anemia was relieved and the ataxia became less marked. Reduction of yeast caused a recurrence of the blood and neurologic changes which were again relieved by an increase in yeast. Eventually secondary infections and extreme ataxia developed, and death occurred.

The growth curves are represented by the interrupted lines. *N.S.* beside an arrow pointing upwards indicates the time of onset, or increase of signs of ataxia. *N.S.* beside an arrow pointing downwards indicates a decrease of ataxia. *P* refers to total inability to walk. *L.E.* indicates the time at which liver extract was injected.

next day, then to 8 per cent, and subsequently more gradually, as the red cell count increased to 7.37 millions and the mean corpuscular volume decreased to 51 c.μ., the mean diameter to 5.10 μ. The stools became normal in consistency and in number and the pig's condition had obviously improved. Two weeks after ataxia was first noticed, it was observed to be much less marked than it had been. The quantity of yeast was reduced again and the anemia as well as the ataxia became more pronounced. A highly concentrated extract of liver (Lederle) was injected, 1 cc. daily for 9 days. The reticulocytes fluctuated between 1.2 and 4.8 per cent but the anemia was unchanged. The animal finally was unable to walk and ulcers in the skin and subcutaneous abscesses formed on the legs, probably as the result of the paralysis and loss of sensation. The quantity of yeast was increased to 6 and, later, 9 grams per Kg. Reticulocytes increased to 7.8 per cent on the sixth day thereafter and the red cell count increased, slowly, from 6.43 to 8.70 millions, the volume of packed red cells from 34.5 cc. to 40.5 cc., while the mean corpuscular volume decreased from 54 to 47 c.μ., the mean diameter from 5.55 to 5.16 μ., and fewer macrocytes remained in the smear. Some improvement in the gait occurred but the animal finally died of bronchopneumonia and empyema. The bone marrow, including that of the shaft of the femur was red and moderately hyperplastic.

No yeast was given 4-80 when the anemia was severe, and death occurred. No infection was found at autopsy. The bone marrow was hyperplastic.

In 4-82 the red cell count decreased to 4.74 millions, the volume of packed red cells to 30.5 cc., the mean corpuscular volume increased from 52 to 65 c.μ., (figure 2) while the mean diameter increased from 5.44 to 5.94 μ. and the blood smear resembled that of 4-79 at the stage of severe anemia. There was diarrhea and moderate ataxia. Administration of yeast (3 gm. per Kg. daily) was followed by a reticulocyte increase to 12.6 per cent on the ninth day following, the red cell count increased to 7.00 million, the volume of packed red cells to 37.0 cc., and the mean corpuscular volume decreased to 53 c.μ., the cell diameter to 5.60 μ. The diarrhea was relieved but the ataxia persisted. As shown in figure 2, decrease in the quantity of yeast was followed by increase of anemia, the mean corpuscular volume rising

again to 60 c.μ. Ataxia became more marked. Administration of yeast was followed again by an increase of reticulocytes to 8.8 per cent although this did not occur until the thirteenth day thereafter. The anemia decreased once more and the mean corpuscular volume decreased to 51 c.μ. At this point, abscesses developed in the hind legs. The general condition of the animal became worse and it was unable to get about at all. A crude liver extract (Campolon) was injected, 5 cc. daily for 19 days. No improvement occurred and death finally ensued. In addition to a number of abscesses on the extremities, lobular pneumonia was found. The bone marrow was moderately hyperplastic.

Gastric Secretion. Achlorhydria was frequently observed in the experimental animals. We are not inclined to attach significance to this finding, however, because failure to find free hydrochloric acid in the gastric secretion following the injection of histamine was not infrequently followed by the discovery of free acid at another examination a few days later. This was as true of the control group of pigs as of those given deficient diets.

Sections of different portions of the stomach, the duodenum and ileum failed to show any lesions except in 4-87, as already mentioned. A few round cells and plasma cells as well as some eosinophils were seen in the mucosa of the colon, especially in those animals in which diarrhea developed and persisted until death. There were no gross or microscopic lesions in the mucosa of the tongue.

Icterus Index. The icterus index of the blood of the control animals was quite constantly between 4 and 6. That of the experimental animals was the same.

No changes were observed in the number of leukocytes other than the leukocytosis which frequently developed in association with infection. As already stated, when anemia was well marked, the platelets appeared to be reduced in number in the blood smears and giant forms were seen.

Antianemic Effectiveness of Basal Diet and of Yeast. The effectiveness of the constituents of the basal diet in producing a reticulocyte response in cases of pernicious anemia, when given alone or after mixture with normal human gastric secretion, was tested in two patients. In patient E. M., the dietary constituents were incubated

with normal gastric juice at pH 3 for two hours, the mixture being neutralized just before feeding. About 70 grams of casein and 150 cc. of gastric juice were given daily in this way. No reticulocyte response occurred.

Castle and Ham (40), however, have recently concluded that incubation of the gastric juice mixture may lead to a decrease in the activity of the intrinsic factor of the gastric secretion. In the second patient (B. A.), therefore, the procedure recommended by these authors was followed. The patient was first given washed casein, 200 grams, as well as washed lactalbumin, 60 grams, daily for 10 days. In the succeeding period of 10 days she was given a daily average of 122 grams casein and 35 grams lactalbumin, mixed with an average of 143 cc. normal human gastric juice. The highest level reached by the reticulocytes during the entire 20 days was 3.4 per cent on the nineteenth day, while the red cell count remained stationary. Subsequently this patient, whose complete record is published elsewhere (1), was shown to respond to yeast given orally. In the last mentioned publication, evidence is presented which demonstrates the antianemic effectiveness of the yeast used in the present experiments with pigs.

DISCUSSION

It seems clear from these experiments that, under the dietary conditions in which they were carried out, some unidentified substance in brewers' yeast is necessary for normal blood formation in the pig. When large amounts of yeast were given, although some fluctuations took place, the number of red corpuscles remained at a high level and no significant morphologic changes occurred, even in animals given reduced amounts of the basal diet ("inanition controls"). When the quantity of yeast was reduced, anemia usually developed. The severity of the anemia varied greatly but it was always characterized by the appearance of macrocytes, polychromatophilia, Howell-Jolly bodies, nucleated red corpuscles and some increase in reticulocytes. The extent of morphologic change varied according to the severity of the anemia. In most instances these changes in the size of the erythrocytes were not sufficiently great to cause a significant increase in the mean volume or mean diameter of the red corpuscles but in several animals an increase in the average size of the cells was observed.

When the anemia was severe the number of blood platelets seemed to be somewhat reduced and unusually large platelets were seen. Leukopenia did not occur. These changes were not accompanied by hyperbilirubinemia nor by significant changes in gastric secretion.

An increase in the quantity of yeast fed was followed, in the four animals in which this type of experiment was carried out, by an increase of reticulocytes which reached its peak within six to thirteen days. In two pigs such an effect was observed twice. The increase in reticulocytes was not as regular as is observed in patients with pernicious anemia receiving specific therapy, but, as in the latter disease, it was followed by a decrease of reticulocytes and, in all but one pig, by the partial or complete disappearance of anemia.

The development of anemia was accompanied by hyperplasia of the bone marrow and in one instance foci of extramedullary blood formation were observed in the spleen. This hyperplasia was clearly not related to the presence of infections at the time of death but was correlated with the degree of anemia, being greater when the anemia was more marked. The hyperplastic bone marrow contained many immature nucleated red cells. While these cells have the appearance of "megaloblasts", their positive identification as such cannot be made at this time.

The anemia seems to have been due largely to the lack of something in yeast other than the vitamins, thiamin, riboflavin or nicotinic acid. This does not deny the possibility, however, that these vitamins have some effect on blood formation. In two pigs (4-60 and 4-87) administration of thiamin when "yeast" deficiency had occurred, was followed not only by an improved growth-rate but by a temporary, moderate relief of the anemia. This effect was observed twice in one of these animals. In two other pigs (4-63 and 4-64) the progress of the anemia ceased when thiamin and riboflavin were administered. Of all the pigs, anemia was least marked in the group given these two supplements.

In a number of instances severe anemia was observed in pigs whose growth and general state of nutrition was poor. It does not appear, however, that the effect of "yeast" deficiency on the blood is through impairment of general nutrition, for anemia did not develop in 2 pigs given half as much food as the rest ("inanition controls") whereas

well marked anemia was observed in animals given thiamin, riboflavin and nicotinic acid, whose growth was very good (group E). In these pigs, in fact, anemia of severe degree developed in each, in striking contrast with the irregularity with which it was observed in the other groups. One gained the impression that in these animals growth and general nutrition were so good that bodily requirements were great enough and life persisted long enough for well marked deficiency of a substance required for blood formation to become apparent.

The identification and synthesis of several of the components of the B complex offers hope that it may be possible to discover the factors in yeast which are concerned in blood formation. Literature has already been cited which describes the development of anemia in pigs, monkeys, rats, dogs and pigeons, as the result of feeding diets low in their content of the vitamin B complex. This anemia has been irregular in its development and the type of morphologic change in the red corpuscles has not always been the same. This irregularity can be explained in part by the multiple nature of the deficiency produced in the majority of the dietary studies which have been made, for purified vitamins were not available at that time. Death or impairment of hematopoietic function may result from deficiency of one substance before the effect of the lack of another factor is evident. Irregularity in the type of anemia produced may thus be explained. Another source of difficulty in studies of this kind is the readiness with which animals lacking yeast develop infections. These may alter the character of the anemia and may inhibit the response to therapeutic agents. Our own experience certainly supports this view.

The changes noted in a number of the pigs were similar in some respects to those found in pernicious anemia. As in the latter disease, defects in the nervous system developed, as well as anemia. Our more recent studies have amply confirmed the preliminary report (2) in which a description was given of the changes in the posterior columns of the spinal cord, the peripheral nerves and the dorsal root ganglion cells. The lesions in the nervous system occurred regularly and were, in fact, much more striking than the changes in the blood. In contrast with the findings in pernicious anemia, leukopenia was not observed nor was there evidence of hyperbilirubinemia or persistent achlorhydria. Furthermore, the injection of liver extracts in two pigs

was without effect. One of these extracts was a highly concentrated one. Its failure to cause a reticulocyte response corresponds to the experience of Wills, Clutterbuck and Evans (41) who found highly refined liver extracts valueless in the cure of macrocytic anemia produced in monkeys. These investigators found crude liver extracts effective. Such an extract was given to one of our pigs but this was done at a time when a severe infection had already developed and thus the conditons for response were not favorable. It is evident that the relation, if any, of the observations described in this report to the disease pernicious anemia, is a matter which cannot be answered by the evidence at hand.

SUMMARY

1. Pigs were weaned at 10 to 23 days of age and placed on a diet consisting of casein, lard, cod liver oil, a mineral mixture, ascorbic acid and yeast. When satisfactory growth had been established the quantity of yeast given some of the animals was gradually reduced and it was replaced by thiamin chloride, riboflavin and nicotinic acid in various combinations.

2. Observations on the blood, gastric secretion and bone marrow of 21 pigs are recorded.

3. Anemia did not develop in 6 pigs given 3 or more grams of yeast per Kg. body weight, but it occurred in all but one of those given smaller amounts of yeast:

(a) Of four pigs given thiamin chloride as the only supplement in place of yeast, slight anemia occurred in three, severe anemia in one. In two of these pigs marked, though temporary, relief of anemia followed the injection of thiamin.

(b) Of five yeast-deficient pigs given thiamin chloride and riboflavin, slight anemia occurred in three, little or none in two.

(c) Of three yeast-deficient animals given thiamin chloride and nicotinic acid, anemia developed in all and it was severe in one.

(d) Of three yeast-deficient animals given thiamin chloride, riboflavin and nicotinic acid, severe anemia occurred in all.

4. The anemia was characterized by the presence of macrocytes, polychromatophilia, Howell-Jolly bodies and nucleated red cells. In most instances no significant change in the mean size of the red

corpuscles was observed but in several instances macrocytic anemia occurred.

5. In four animals in which such therapy was attempted, administration of yeast was followed by reticulocytosis and partial or complete relief of anemia. In one of these animals a purified liver extract had no effect. In another a cruder extract was used without result but it is possible that the presence of an infection inhibited the response.

6. Hyperplasia of the bone marrow was observed but persistent achlorhydria and hyperbilirubinemia did not occur.

Thiamin chloride was kindly furnished by Mead Johnson and Company and Winthrop Chemical Company, Inc., and ascorbic acid and riboflavin by Merck and Company, Inc. Technical assistance was given by William Thaler, Stewart Humphreys, Miss Betty Karns, Thomas Shafferman and John Drasal. Photographs by Milton Kougl.

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PLATE I

FIG. 1. Blood of pig 4-79 at 89 days of age when there was no anemia. Oil immersion (× 1170).

FIG. 2. Blood of same pig at 141 days of age when there was anemia and macrocytosis. Note Howell-Jolly bodies. Oil immersion (× 1170).

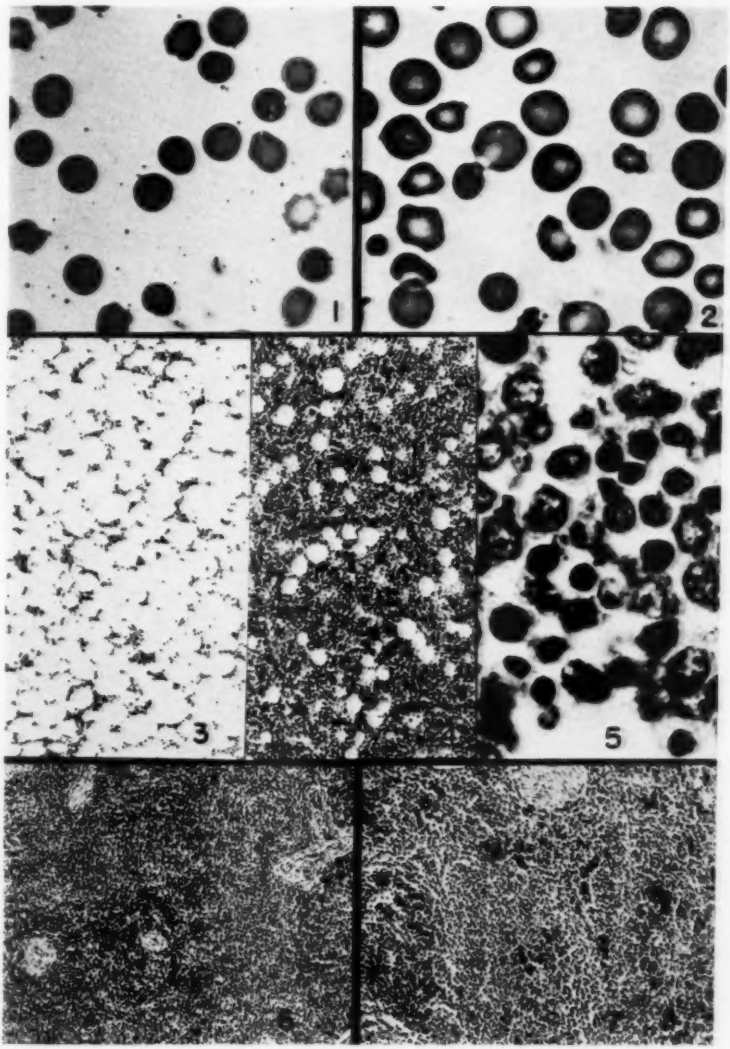
FIG. 3. Femur marrow of control pig (× 90).

FIG. 4. Femur marrow of anemic pig (4-97), showing hyperplasia (× 90).

FIG. 5. Femur marrow of same pig (× 700). Note scarcity of mature cells.

FIG. 6. Spleen of pig 4-97 (× 90). Normal.

FIG. 7. Spleen of pig 4-94 (× 90). Many foci of extramedullary blood formation.



UNIV. OF MICH.

DIETARY PROTEIN AND THE REGENERATION OF SERUM ALBUMIN

III. The Potency Values of Egg White, Beef Liver and Gelatin

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The first paper of this series (1) dealt with the principles underlying a new method for evaluating the potency of various food proteins with respect to their ability to promote the formation of serum albumin. In a second paper (2) the potency values of beef serum, beef muscle, and casein were compared. In the present communication potency values for egg white, beef liver, and gelatin will be reported together with a statistical comparison of the relative merits of the six foods which have so far been examined.

The method of conducting the assays involves the use of dogs in which deficits in serum albumin have been produced by feeding a basal low protein diet for a standard period of 3 weeks. The regeneration of albumin is measured during the ensuing week when the diet is composed of a mixture of the basal diet and the food protein to be tested. The mixture is in such proportions that each animal receives daily 80 calories and 2.5 grams of protein per kilo. The *assay value* of a food is calculated as 0.15 plus the figure which represents, in grams per 100 cc., the rise in albumin concentration in the serum during the week in which the food was fed. The constant, 0.15, is to be regarded as an allowance for maintenance; it corresponds to the fall in albumin which would have been expected if no supplementary protein had been incorporated in the diet. Since replicate measurements of the same food with different animals disclose a fairly wide range of biological variation among the assay values, the *potency value* of a food is determined by averaging a number of assay values from experiments with different animals.

DIETS

The composition and preparation of the basal low protein diet has been described in an earlier paper (3). It is necessary to discuss here only the test foods.

Egg white. Fresh eggs were boiled for from 15 to 20 minutes; the whites were separated and chopped with a knife into coarse particles. This material was stored in the ice-box in tightly covered cans to prevent loss of moisture until needed in the feeding experiments, a period of about 3 days. The required quantity of egg white was weighed separately for each dog and mixed with the basal diet just before feeding. Analyses of the coagulated egg white disclosed an average protein content (6.25 × N) of 12.3 per cent. The regeneration diet was a mixture of 53.4 grams of basal diet and 16.62 grams of the egg white per kilo of dog weight. In this mixture 81.8 per cent of the total dietary protein is furnished by the egg white.

Beef liver. The coarse fibrous tissue surrounding the large branches of the portal system was removed from fresh beef livers and the remaining portion chopped in a meat grinder. This material was stored in the ice-box in covered cans until just before feeding when the required amount was mixed with the basal diet. Analyses of the ground liver disclosed an average protein content (6.25 × N) of 21.6 per cent. This figure is within the range of values for beef liver protein tabulated by Atwater and Bryant (4). The same tables give a fat content from 3.3 to 5.7 per cent; the regeneration diet was calculated on the basis of 4.5 per cent fat. The composition of the diet in terms of amount fed per kilo was: basal diet, 50.3 grams; beef liver, 9.59 grams. With this mixture 82.8 per cent of the total dietary protein is in the form of liver.

Gelatin. A commercial brand of gelatin was purchased in granulated form. The required quantity was soaked in a small amount of water for about 10 minutes before mixing with the basal diet and feeding. Analyses of the gelatin disclosed an average protein content (6.25 × N) of 97.6 per cent. Two assays were run in which the regeneration diet furnished the usual 2.5 grams of protein per kilo; in terms of amount fed per kilo this diet contained: basal diet, 53.4 grams; gelatin, 2.09 grams. In two other assays the regeneration diet furnished 5 grams of protein and 80 calories per kilo; the components of this mixture were: basal diet, 45.5 grams; gelatin, 4.72 grams. These two diets furnished respectively, 81.8 per cent and 92.1 per cent of the total dietary protein as gelatin.

EXPERIMENTAL FINDINGS

The data obtained in eleven assays of egg white and in eleven assays of beef liver are summarized in Tables I and II. The potency value (average assay value) of egg white with its probable error is 0.616 ± 0.023; the potency value of beef liver is 0.445 ± 0.025.

The four assays of gelatin which were made at two different levels of protein feeding are brought together in Table III because regardless

TABLE I

Assays of Egg White for Albumin Formation

DOG	SERUM ALBUMIN PER 100 CC.					ASSAY VALUE
	Initial	Depletion	Regeneration	Loss	Gain	
	gram	*gram*	*gram*	*gram*	*gram*	
2-14	3.51	2.41	2.64	1.10	0.23	0.38
5-31	4.03	2.96	3.27	1.07	0.31	0.46
3-08	3.82	2.52	2.95	1.30	0.43	0.58
3	3.84	2.85	3.29	0.99	0.44	0.59
3-36	4.24	3.28	3.75	0.96	0.47	0.62
7-31	3.47	2.43	2.90	1.04	0.47	0.62
Bol	3.47	2.19	2.71	1.28	0.52	0.67
1-55	3.64	2.59	3.13	1.05	0.54	0.69
7-27	3.56	2.21	2.77	1.35	0.56	0.71
2-05	3.28	1.89	2.46	1.39	0.57	0.72
1-69	3.25	2.17	2.75	1.08	0.58	0.73

Potency value: 0.616 ± 0.023 (P.E.).

TABLE II

Assays of Beef Liver for Albumin Formation

DOG	SERUM ALBUMIN PER 100 CC.					ASSAY VALUE
	Initial	Depletion	Regeneration	Loss	Gain	
	gram	*gram*	*gram*	*gram*	*gram*	
9-52	3.30	1.89	2.01	1.41	0.12	0.27
3	3.89	2.98	3.13	0.91	0.15	0.30
2-66	4.52	3.13	3.36	1.39	0.23	0.38
2-14	3.52	2.04	2.28	1.48	0.24	0.39
8-04	3.82	2.96	3.22	0.86	0.26	0.41
3-36	4.15	3.19	3.46	0.96	0.27	0.42
2-07	3.84	2.43	2.70	1.41	0.27	0.42
5-31	4.11	2.87	3.20	1.24	0.33	0.48
1-55	4.19	2.99	3.36	1.20	0.37	0.52
9-51	3.89	2.97	3.47	0.92	0.50	0.65
7-31	3.74	2.52	3.02	1.22	0.50	0.65

Potency value: 0.445 ± 0.025 (P.E.).

of the level of gelatin in the diet, there is no evidence that this protein can be used at all to supply the needs of the plasma. Calculation of

the assay values in these experiments represents a modification of the usual procedure. The modification, which was discussed in a previous paper (1), is introduced because the number of assays is small and it is desired to know as accurately as possible whether or not gelatin possesses any value. Since the decline in albumin concentration during a fourth week of feeding the basal diet is influenced by the actual level reached at the end of 3 weeks, it is possible in each assay experiment to estimate the decline which would have been expected

TABLE III

Assays of Gelatin for Albumin Formation

DOG	PROTEIN IN DIET DURING REGENERATION	SERUM ALBUMIN PER 100 CC.					ASSAY VALUE
		Initial	Depletion	Regeneration	Loss	Gain	
	gram per kg.	*gram*	*gram*	*gram*	*gram*	*gram*	
7-27	2.5	3.86	2.54	2.10	1.32	−0.44	−0.26
8-03	2.5	3.72	2.05	1.85	1.67	−0.20	−0.09
7-29	5.0	3.80	2.80	2.54	1.00	−0.26	−0.04
5-31	5.0	4.38	3.19	2.93	1.19	−0.26	+0.02

Potency value: -0.093 ± 0.041 (P.E.).

if the test protein had not been added to the diet. The estimate is made from the regression equation:

$$\triangle_A = 0.144A - 0.18$$

in which $\triangle_A$ is the expected decrement during the fourth week and A equals the albumin concentration after 3 weeks, in grams per 100 cc. of serum. In each experiment with gelatin the figure representing the estimated value of $\triangle_A$ in grams per 100 cc. was used instead of the constant, 0.15, as an allowance for maintenance in calculating the assay value. The potency value of gelatin, which is less, though not significantly, than zero, is -0.093 ± 0.041. If the routine procedure had been used in computing the assay values, the potency value would have been recorded as -0.140 ± 0.035.

DISCUSSION

In the left hand column of Table IV the potency values given in this paper, together with those reported previously, are arranged in

order of magnitude. Since the accuracy of the potency values is determined both by the number of assays (indicated in the table by figures in parenthesis) and by the variability of the assay values, it is necessary that these factors receive consideration before the meaning of a difference between two potency values can be appraised. Accordingly the probability integral has been computed for the difference between the potency values of each two of the six foods; the values of the integral are also given in Table IV. In these calculations we have followed the method of Student as elaborated by Fisher (5); values of the integral corresponding to computed values of t have been taken from the tables of Treloar (6). According to prevalent custom values of the probability integral of 0.05 or less are indicative of a significant difference between the groups being compared; values of

TABLE IV

		GELATIN	CASEIN	BEEF LIVER	BEEF CHUCK	EGG WHITE
Beef serum (11).....	0.801	0.0001	0.0001	0.0003	0.0007	0.042
Egg white (11).....	0.616	0.0001	0.0003	0.0027	0.0058	
Beef chuck (12).....	0.475	0.0001	0.098	0.42		
Beef liver (11).....	0.445	0.0001	0.38			
Casein (12).....	0.388	0.0001				
Gelatin (4).....	−0.093					

0.01 or less are highly significant. Beef serum, which yielded a higher potency value than any of the other foods, is without question more effective in promoting albumin synthesis than casein, beef liver, or beef muscle. The difference between beef serum and egg white is barely significant. Egg white is secure in second place and there can be no doubt concerning the absolute inferiority of gelatin. However, a significant difference between beef muscle, beef liver, and casein has not been demonstrated.

The meaning of the negative potency value assigned to gelatin deserves brief comment. In three of the four animals which were used for assaying this protein, the level of albumin in the serum at the end of the regeneration period was less than would have been expected if no protein at all had been added to the basal diet. The result, which suggests that gelatin promotes destruction rather than

synthesis of serum albumin, cannot be stressed since the experiments are few and the negative character of the potency value has not been established with statistical certainty. Nevertheless, it is of interest to observe that gelatin contains a high percentage of glycine (7) and that glycine is known to possess a strong specific dynamic action (8). By raising the output of energy it is therefore possible that gelatin may stimulate the destruction of body protein without being able to furnish the building stones required for repair. That the inability of gelatin to stimulate the production of plasma protein is actually the result of a lack of essential amino acids, has been demonstrated recently by Madden, Noehren, Waraich, and Whipple (9). These authors have shown that gelatin supplemented by cystine and tryptophane or by cystine and tyrosine is almost as effective as beef serum for this purpose.

The potency values discussed in this paper can be compared with similar data which have been reported by Whipple and associates (10). These investigators have utilized the technic of plasmapheresis to measure the efficiency of a number of food proteins in fabricating total protein in plasma. Their results are given in the form of *potency ratios* which refer to the number of grams of a food necessary for the formation of one gram of plasma protein. It will be observed that the more potent the food the lower will be the potency ratio, a relationship which is the inverse of that which holds for the potency values in this paper. The protocols which accompany Whipple's reports supply data concerning the relative amounts of albumin and globulin in the plasma so that it is possible to calculate for his experiments potency ratios which refer to the formation of albumin alone. We have made such calculations for the six foods under consideration. The results are given in Table V. It is important only to point out that the order of efficiency is the same for all of the six foods. The relative magnitudes of the values cannot be compared satisfactorily since the methods of assay were entirely different.

Finally, it is of interest to compare the potency values for albumin formation with what is known concerning the efficiency with which various foods are utilized to protect and augment the proteins of the entire body in metabolism. From experiments chiefly on the slowly growing rat Mitchell, and others who have followed his methods, have

ascribed *biological values* to the more important food proteins. The biological value refers to the percentage extent to which the digestible protein of a food meets the requirement of the body. Since utilization depends also upon digestibility, the *net value* of a food represents the product of the biological value and the percentage digestibility. In Table VI these values are given for the six foods considered in this

TABLE V

Relationship between (a) the Potency Ratio for Synthesis of Total Plasma Protein (Whipple and Associates), (b) the Potency Ratio for Synthesis of Serum Albumin (Computed from the Data of Whipple), and (c) the Potency Value for Forming Serum Albumin

SOURCE OF FOOD PROTEIN	POTENCY RATIO, WHIPPLE AND ASSOCIATES		POTENCY VALUE, WEECH AND GOETTSCH
	Total protein	Albumin	
Beef serum	2.6	4.8	0.801
Egg white	5.9	10.8	0.616
Skeletal muscle	5.7	12.4	0.475
Liver	6.6	13.8	0.445
Casein	10.0	24.2	0.388
Gelatin	13.6	27.2	−0.093

TABLE VI

Relationship between (a) the biological value, digestibility, and net value of food proteins, and (b) the potency value for forming serum albumin

The foods are arranged in descending order of the potency values for albumin formation

	BIOLOGICAL VALUE	COEFFICIENT OF DIGESTIBILITY	NET VALUE
Beef serum	59	82	48
Egg white	83	100	83
Beef muscle	69	96	66
Beef liver	77	90	69
Casein	66	89	59
Gelatin	0	?	0

paper. We are indebted to H. H. Mitchell for calling our attention to the work of Arsenjew and Chlebnikow (11) in Moscow who appear to be the only investigators to have measured the biological value of serum protein. The measurements which were made in chickens yielded a biological value of 78 at 20 days of age and of 59 at 45 days of age. The lower of these values has been entered in the table since in other experiments it was shown that beef serum is inferior to casein in promoting growth in weight. The value for casein in Table VI was

obtained in rats by Morgan (12); the remaining figures were taken from data assembled by Mitchell and Hamilton (13). Except for beef serum the agreement in order of efficiency between either biological value or net value and the potency value for albumin formation is reasonably close. The values for beef muscle and beef liver are in reverse order in the two series but in both series the values are close together and the difference between them too small to be significant. Beef serum, however, is clearly out of line, with a high potency value for forming albumin and a low biological value.

SUMMARY

A method previously described for establishing the efficiency of food proteins in forming serum albumin is outlined. Eleven assays of egg white, eleven assays of beef liver, and four assays of gelatin are reported. The data yield the following potency values for albumin formation: egg white, 0.616 ± 0.023; beef liver, 0.445 ± 0.025, gelatin, -0.093 ± 0.041. Similar potency values have already been reported for three other foods: beef serum, 0.801 ± 0.053; beef muscle, 0.475 ± 0.021; casein, 0.388 ± 0.027.

The data which gave the potency values for the six foods have been examined by conventional statistical procedure to establish the reliability of the order of efficiency. Beef serum and egg white are clearly more potent in forming serum albumin than any of the other four foods. The difference between beef serum and egg white is barely significant. There is no doubt concerning the inferiority of gelatin. However, a significant difference between beef muscle, beef liver, and casein has not been demonstrated.

The same order of efficiency in promoting albumin formation can be computed for the six foods from data previously supplied by Whipple. Although a relationship is evident between the potency values for albumin formation and the biological values of the proteins of the foods, it is significant that beef serum possesses a high potency value and a relatively low biological value.

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THE PATHOGENICITY OF AVIRULENT PNEUMOCOCCI FOR ANIMALS DEPRIVED OF LEUCOCYTES

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The pathogenicity of a bacterium, *i.e.*, its power to damage the tissues of a given host, resides primarily in its ability to propagate itself progressively in the tissues of the host; for, obviously, damage to the body of the host by exotoxins or endotoxins of bacteria which gain entry into the tissues in natural infection can occur only if the bacteria are able to increase to numbers sufficient to liberate an appreciable amount of their toxic products; and the proliferation of the bacteria is also essential for the production of any damaging effects referable to the hypersensitive state, and also for the occurrence of the foreign-body reactions that are produced by various bacteria, such as the tubercle bacilli, which contain substances that the body cannot readily bring into a soluble state. Some bacteria are non-pathogenic for a given host because they are promptly ingested and destroyed by the phagocytes of that host, and are thus prevented from multiplying progressively; others are non-pathogenic for the reason that the physical environment of the tissues of the host (*e.g.*, the normal body temperature) is unfavorable for their proliferation, though they may remain alive for months in the tissues (*e.g.*, mammalian tubercle bacilli injected into cold blooded animals); others, as we have recently shown (3), are able to proliferate in the normal body until the infection, itself, elevates the body temperature to a fever level unfavorable to the proliferation and survival of the bacteria, as a result of which the progress of the infection is promptly terminated; still others are non-pathogenic because the chemical constitution of the tissue fluids of the host fails to provide in utilizable form the nutritive materials that are necessary for the proliferation of the bacteria; but in all cases non-pathogenicity is determined

primarily by the inability of the bacteria to proliferate progressively in the body of the host, whatever the manner of prevention of their growth may be.

As is well known, both the host and the bacterium are variables, either of which can be modified with a resulting change in the degree to which the bacteria will be able to proliferate in the tissues. Thus, the body of a susceptible host may, by immunization, be altered in such a manner that a given pathogenic bacterium can no longer proliferate freely in its tissues and is thereby rendered harmless; or a host that is natively resistant to a particular bacterium that is pathogenic for other species may, by an alteration of its body temperature, be placed in a condition favorable to the free proliferation of that bacterium, which can then readily overwhelm it. On the other hand, leaving the body of the host as a constant, the constitution of a given bacterium that is highly virulent for that host, may be so modified that the bacterium, though able to grow readily *in vitro*, can no longer proliferate freely in the tissues of the host, and thereby becomes harmless even though, in some cases, it may survive for long periods of time in the tissues. Certain attenuated strains of tubercle bacilli illustrate very well this latter situation. Or, finally, a bacterium that has become attenuated to the point of non-pathogenicity can through the alteration of its constitution by appropriate measures, be restored to full virulence with the attendant power of proliferating freely and progressively in the tissues of the host.

The *in vitro* conversion of an encapsulated pneumococcus that is virulent for rabbits, into the non-encapsulated, avirulent form is a familiar example of the situation in which alteration of the constitution of the bacterium profoundly alters its pathogenicity for a fixed host. With the loss of its polysaccharide capsule the pneumococcus loses its high degree of virulence and becomes virtually non-pathogenic. In the tissues of the normal rabbit in which the encapsulated form proliferates freely to overwhelming numbers, the non-encapsulated form is unable to survive and rapidly disappears. *In vitro* and *in vivo* studies readily show that the encapsulated, virulent pneumococci resist phagocytosis, while the non-encapsulated form is rapidly ingested and destroyed by the leucocytes. Phagocytosis, therefore, plays a highly important rôle in the native immunity of the rabbit to

the non-encapsulated pneumococcus. Whether this native immunity can be explained entirely on the basis of phagocytosis has, however, not been previously determined. Previous studies have failed to establish whether or not the conversion of the virulent, to the avirulent form has so altered the bacterium that its ability to survive and to proliferate extracellularly in the tissues has been lost; and it has remained undetermined whether or not the avirulent, non-encapsulated form, if it could survive in the tissues, would be capable of producing the same type of tissue damage that is characteristic of the lesion caused by the virulent, encapsulated organism.

In the course of a study of the factors concerned in native and acquired immunity to various bacteria, it seemed desirable to attempt to learn whether the non-pathogenicity of the avirulent, non-encapsulated pneumococcus is determined solely by its susceptibility to phagocytosis, or whether in the change from the encapsulated to the non-encapsulated form the bacteria undergo other alterations of constitution which render impossible their survival or proliferation in the environment within the animal body. In addition, we wished to learn whether the type of lesion produced by the avirulent non-encapsulated form, if it could be induced to proliferate in the tissues, would differ from that produced by the encapsulated, virulent form. The present experiments were carried out for the purpose of studying these points.

EXPERIMENTS

In order to determine whether avirulent pneumococci are able to proliferate in the tissues if they are left there undisturbed by phagocytes, it was necessary to remove the phagocytic defense factor from the picture. In a previous study of the question of the existence of "tissue immunity" in acquired resistance (1) we had followed the fate of virulent pneumococci in the tissues of the immunized body which had been deprived of leucocytes by treatment with benzol. Precisely the same method of removing the leucocytes was employed in the present study, and since the procedure of benzol treatment and its effects were described in considerable detail in the former paper we shall not repeat it here.

The organism used in the present study was an avirulent pneumo-

coccus dissociated from a virulent, encapsulated strain of Type II. It was bile soluble. No capsule could be detected by the India-ink method or by capsule stains. The colony growth was "rough." In contrast to the encapsulated form, these non-encapsulated pneumococci were promptly ingested by the leucocytes of the normal rabbit both *in vitro* and *in vivo*. Rapid and avid phagocytosis by polymorphonuclear and mononuclear leucocytes was readily demonstrable in pleural exudates following the intrapleural injection of the bacteria, and also in sections of skin following intracutaneous injection. Phagocytosis by the Kupffer cells of the liver was readily observable following injection of the bacteria into the portal vein. In contrast to the virulence of the encapsulated Type II pneumococcus for the rabbit (one ten-millionth of a cubic centimeter of a highly virulent blood broth culture may suffice to kill the animal within 36 hours), we have injected the bacteria from as much as 10 cc. of undiluted heavy blood-broth culture of the non-encapsulated form without fatal effect, and doubtless the rabbit could dispose of even more.

When 0.1 cc. of a blood-broth culture of a virulent, encapsulated Type II pneumococcus is injected into the skin of the upper part of the flank of a rabbit, an oedematous, erythematous lesion appears within several hours. This grows progressively larger, spreading downwards until it reaches the abdominal surface. As it progresses, haemorrhages often make their appearance in the lesion, and these may become extreme and confluent. The blood culture becomes positive after several hours, and the animal dies with a heavy septicaemia within 24 to 36 hours.

In contrast, the intracutaneous injection of the same dose of the non-encapsulated form causes only a small, slightly oedematous and hyperaemic lesion with no haemorrhage, which regresses and disappears. The blood culture remains sterile. Microscopical study of numerous sections through each individual lesion produced by the virulent form shows a steadily progressing inflammation, which becomes more and more intense and extensive up to the death of the animal. The number of pneumococci in the lesion increases continually from the beginning, and after some hours they are present in swarms. None are engulfed by the leucocytes that surround them. In the later stages haemorrhages are often prominent, and there is

necrosis of the corium and of the underlying muscle. In contrast, at the site of injection of the non-encapsulated pneumococci there occurs a localized inflammatory outpouring of fluid and leucocytes which is far less extensive than that caused by the virulent form, and which, having reached its height during the first 24 hours, then undergoes regression. Necrosis of the corium is minimal and is confined to the line of trauma produced by the injection; there is no destruction of the underlying muscles such as is regularly seen at the site of injection of the virulent organisms, though occasionally a single hyalinized fibre may be found; and there is no haemorrhage (Fig. 1). The bacteria can be found only in lesions removed during the first few hours. When the leucocytes appear they begin at once to ingest the bacteria, and very soon no pneumococci at all can be found in the sections stained to reveal them, in contrast to the swarms stained in the tissues infected with the virulent form and removed at the same time.

In the six rabbits that were infected with the avirulent pneumococci after having been deprived of leucocytes by treatment with benzol, the course of the infection was strikingly different from that in the similarly infected controls possessing leucocytes (Table I). When the benzol treatment had reduced the circulating leucocytes to the level

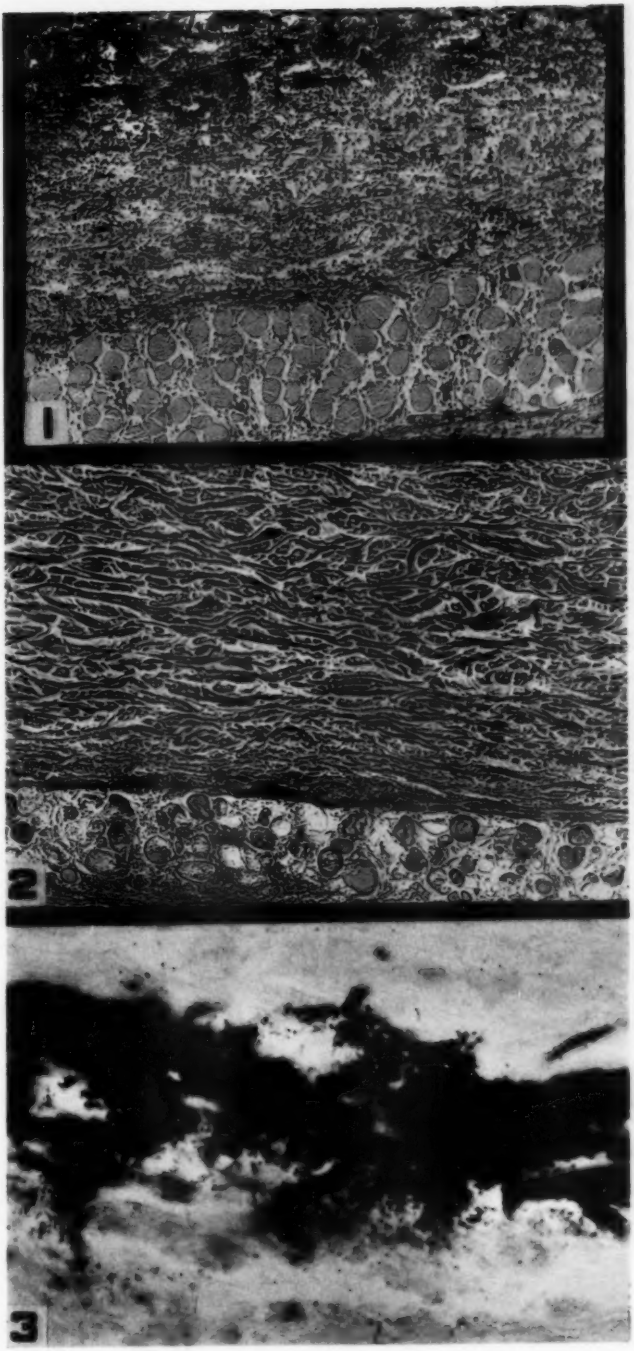
EXPLANATION OF ILLUSTRATIONS

(Photographs by Mr. Milton Kougl)

FIG. 1. Skin of a normal rabbit 58 hours after the injection of 0.1 cc. of a culture of non-encapsulated, avirulent pneumococci. Inflammation is present; the bacteria have all been destroyed; the muscles beneath the corium are normal. Haematoxylin and eosin stain.

FIG. 2. Skin of a leucopenic rabbit 58 hours after the injection of 0.1 cc. of a culture of non-encapsulated, avirulent pneumococci. There is no inflammation; the bacteria have multiplied to enormous numbers, as the bacterial stain in Fig. 3 shows; and there is necrosis of the connective tissue cells of the corium and of the muscles beneath the corium. Some of the muscle fibres are hyalinized, others are partly or completely liquefied. There is haemorrhage between the fibres. Haematoxylin and eosin stain.

FIG. 3. High-power photomicrograph of bacterial stain of the site of infection of animal illustrated in Fig. 2. The solid black areas are masses of bacteria, and individual cocci are visible in the less closely packed areas. Polychrome methylene-blue stain for bacteria.



FIGS. 1–3

that previous experience (1) had shown to be favorable for the production of leucocyte-free lesions, 0.1 cc. of an eight to twelve hour blood-broth culture of the avirulent pneumococcus was injected into the skin of the depilated flank, and the same amount of the same culture was injected at the same time into the skin of normal control rabbits. The sites of infection were carefully observed macroscopically, and were removed for microscopical study at the end of the experiment. Blood cultures were made at intervals during the course and at the end of the experiments. The length of life of the six leucopenic animals following infection was, respectively, 40, 42, 55, 58, 64 and 80 hours. All control rabbits survived. The receding lesions of the controls were removed for histological comparison with those of the leucopenic animals when the latter died.

The macroscopic lesion in each control animal was a slight, non-progressive one—the evanescent slight oedema and hyperaemia described above as the typical result of the intracutaneous injection of the avirulent pneumococcus. In none of the controls did the blood culture ever become positive.

The site of infection in the leucopenic animals was usually less erythematous and oedematous than that in the control animals. In our previous paper (1) we described and commented upon this lesser degree of hyperaemia and oedema at sites of infection in benzol-treated animals. In marked contrast to the slight lesions in the controls, however, there developed in three of the six leucopenic animals progressively spreading macroscopic lesions that were extensively haemorrhagic. The avirulent pneumococcus was recovered from the blood of two of these three animals; from one just before death 64 hours after infection, and from the other at 32 and 44 hours after infection and again shortly after death 36 hours later. The pneumococci recovered from the blood in these animals were in all respects identical with the avirulent ones that were injected into the animals. They were bile soluble, produced typical rough colonies on blood agar, were non-encapsulated, and produced only the characteristic slight reaction when injected intracutaneously into normal rabbits.

The spreading, haemorrhagic macroscopic lesions in the three leucopenic animals described above were completely different from

the slight oedema and hyperaemia characteristic of the effect of the avirulent pneumococcus in the normal animal. Histological study of the lesions revealed at once the reason for the difference, and also made it clear that the lesions in the other three leucopenic animals were progressing toward the same severe end which, undoubtedly, would have been reached had the animals not died. Whether these animals died from the effects of the benzol, or from absorption of toxic products from the bacteria-laden lesions could not be determined. In contrast to the lesions in the control animals, in which leucocytes were present in abundance and in which no pneumococci at all could be found as a result of their prompt ingestion and destruction by

TABLE I

Results of intracutaneous infection with non-encapsulated, avirulent pneumococci in leucopenic and control animals

TYPE OF ANIMAL	NUMBER OF ANIMALS	UNDILUTED CULTURE OF AVIRULENT PNEUMOCOCCUS	CHARACTER OF LOCAL LESION AFTER 24 HOURS				POSITIVE BLOOD CULTURE
			Haemorrhage	Necrosis of muscle	Bacteria at site	Leucocytes at site	
		cc.					
Leucopenic.....	6	0.1	+ (4)	+ (6)	Large numbers to myriads (6)	Few or none (6)	+ (2)
Control.....	6	0.1	0 (6)	0 (6)	None (6)	Large numbers (6)	0 (6)

The figures in parentheses represent the number of animals showing the conditions listed in the various sections.

the phagocytes, in the lesions of the leucopenic animals few or no leucocytes were present and pneumococci were present in swarms (Figs. 2 and 3). In the lesions into which only a very few leucocytes, or none at all, had immigrated, the pneumococci were present in extraordinary numbers, forming solid masses that occupied the tissue spaces of the corium and extended in dense clouds into the spaces between the underlying muscles. In two lesions in which leucocytic emigration was more evident, though still trivial in comparison with that in the controls, the number of pneumococci was decidedly smaller; but even here their proliferation was unmistakable, for while the scattered leucocytes had been able to dispose of many of the organisms, many had escaped contact with the phagocytes and continued to

proliferate. In one of the three lesions in which haemorrhage was not seen macroscopically, microscopic haemorrhages were evident deep in the tissues; and in all six of the lesions in the leucopenic animals there was marked necrosis of the muscles immediately underlying the lesion. At a distance from the lesion the muscles beneath the corium were always normal.

DISCUSSION

The study of the lesions in these animals makes it impressively clear that non-encapsulated, avirulent pneumococci are able to grow freely in the tissues of the rabbit if they are unmolested by the phagocytes. The change from the virulent, encapsulated, non-phagocytizable form, to the avirulent, non-encapsulated phagocytizable form, therefore, does not entail any alteration of the microörganism that prevents its survival or multiplication in the animal body if the activity of the phagocytes is prevented. Spared from the attack of the phagocytes the avirulent organisms behave in the same manner as do virulent ones, *i.e.*, they multiply progressively to overwhelming numbers, and invade the blood stream and establish a septicaemia. Furthermore, in the absence of the leucocytes they produce a local lesion that has the essential destructive features (necrosis, haemorrhage) characteristic of a severe lesion produced by the virulent, encapsulated form. It is therefore clear that the factor that determines the virulence of the bacterium (*i.e.*, the polysaccharide capsule that prevents phagocytosis) is not essential for the production of the haemorrhagic, necrotic type of lesion, characteristic of the action of virulent pneumococci in the normal body. This action is a property of the somatic portion of the pneumococcus, and the change in the organism resulting in the failure to produce a capsule does not alter the ability of the remaining somatic portion to produce the characteristic tissue damage.

It may be noted that it required a considerably longer time for the avirulent pneumococci to establish a septicaemia in the leucopenic animals than it does for virulent pneumococci to do so whether in leucopenic or in normal animals. The earliest appearance of the septicaemia in the former case was at 32 hours, whereas the blood culture ordinarily becomes positive during the first 12 hours in both

normal and leucopenic rabbits infected with the same dose of virulent pneumococci. There are at least two possible reasons for the delayed appearance of septicaemia in the case of infection with the avirulent form. The delay in septicaemia is certainly not due to a scarcity of bacteria in the local lesion, for the avirulent pneumococci were found in larger numbers in the local lesions of the leucopenic rabbits without septicaemia than in lesions produced by virulent pneumococci at the time the blood culture becomes positive. The delay in the appearance of septicaemia in the case of the avirulent form may be due in the first place to the fact that these bacteria have a tendency to grow in chains and clumps in the tissues in contrast to the virulent pneumococci which separate from each other as they divide, and therefore drift more readily through the tissue spaces. The growth of the avirulent form in the tissues resembles that of the virulent form in a body that possesses a rather weak agglutinating antibody. Our previous studies (1) (2) brought to light the fact that in acquired immunity the agglutinating antibody acts to inhibit the free drift of the virulent bacteria through the tissues and into the lymphatics and blood stream by causing them to adhere to one another and to the tissues as they divide, thus producing clumps and masses which cannot drift readily through the tissue spaces. This function of the immune antibody in preventing the spread of infection is a highly important one. A cohesive process, different in mechanism and in degree, but similar in effect, appears to play a rôle in delaying the spread of the avirulent pneumococcus in the normal body.

Finally, there is one other factor which undoubtedly plays a very important rôle in delaying the appearance of septicaemia. This factor is the persistence of phagocytic cells in contact with the blood stream in the leucopenic animals. Not only do some phagocytic leucocytes remain in the circulation in the benzol treated animals, but, as we pointed out and illustrated in our previous paper (1), the phagocytic, intravascular macrophages ("reticulo-endothelial cells") of the liver and spleen are more resistant to the action of benzol than are the circulating polymorphonuclear leucocytes, and these macrophages, particularly those in the liver (Kupffer cells), remain in considerable numbers and in active condition when the circulating polymorphonuclears are reduced to a small fraction of their normal number. As

is well recognized, these fixed macrophages play a very important rôle in keeping the circulation free from any foreign or abnormal particulate matter that they are able to phagocytize, and they avidly phagocytize non-encapsulated pneumococci. It is not to be wondered at, therefore, that it is difficult to establish a septicaemia with these readily phagocytizable bacteria. Indeed, the wonder is that, having to run the gauntlet of those extraordinarily efficient phagocytes, the avirulent pneumococci ever are present in the circulation in numbers sufficient to be detected by blood culture. It may be mentioned that in our previous study of the reaction of leucopenic animals to virulent pneumococci (1) it was found that a much longer time was required for the appearance of septicaemia in immunized leucopenic animals than in non-immune leucopenic animals. The rôle played by the reticulo-endothelial phagocytes of the liver and spleen in delaying the appearance of septicaemia in the immunized animals was pointed out in that paper, and the ability of these cells in the immunized benzol-treated animals to phagocytize the virulent pneumococci that were opsonized by the immune antibody was illustrated in photomicrographs. The activity of these cells in delaying septicaemia by removing virulent, encapsulated pneumococci from the circulation of an animal that possesses opsonizing antibody is quite analogous to their activity in this regard in the present experiments, in which the form of pneumococcus used (non-encapsulated) is readily phagocytized by these cells in the absence of antibody.

Whether there are any additional factors which may influence the rate of appearance of septicaemia in the present experiments we are at present unable to say. A comparison of the number of avirulent pneumococci in the tissues of the leucopenic animals at various intervals of time following infection, with the number of virulent pneumococci that, in our previous study (1), were present in the tissues after the same lapse of time, gives the impression that the multiplication of the latter proceeded more rapidly than that of the former; but since *in vitro* studies of rate of growth in fluid media, by means by plating and colony counts at various intervals, showed that the virulent strain proliferates even *in vitro* more rapidly than does the avirulent one, we are unable to ascribe the difference in rate of proliferation *in vivo* to an effect of the living body or its fluids.

The present experiments provide a clear illustration of the interdependency of the two variables, bacterium and host, in determining the occurrence of disease. Starting with a virulent pneumococcus that has the ability to proliferate freely in the normal rabbit and to produce a characteristic progressive local lesion and septicaemia, the bacterium is deprived of its capsule and so rendered avirulent, and powerless to cause either a progressive local lesion or septicaemia in the normal host; for without its capsule it can be readily phagocytized and destroyed by the leucocytes. Then the other variable, the host, is altered by depriving it of its leucocytes; and now the avirulent bacteria are once again able to proliferate freely in the tissues and to produce the characteristic progressive lesion and septicaemia.

Since, if protected from the phagocytes, and so permitted to proliferate in the tissues, this avirulent bacterium can exhibit markedly destructive and invasive properties, it is not inconceivable that some of the many bacteria that are non-pathogenic for all animals may be so only because their proliferation is prevented by the phagocytes; and one is tempted to wonder what disease processes such bacteria might produce were their proliferation permitted by removing the phagocytic defense. We have so far made only a single attempt in this direction, and that without success. A non-pathogenic bacillus derived from the air, that grew well on ordinary media at body temperature, was introduced into the skin of leucopenic animals but, although no leucocytes emigrated at the site, the bacillus failed to grow at all. It is evident that some bacteria are non-pathogenic simply for the reason that the tissue fluids of the animal body are such that they do not permit the growth of the bacteria, irrespective of any activity of the leucocytes.

SUMMARY

When non-encapsulated pneumococci, which are non-pathogenic (avirulent) for normal rabbits, were introduced into the tissues of rabbits deprived of leucocytes, they exhibited the qualities of virulent pneumococci, *i.e.*, they multiplied to enormous numbers and produced a progressive local infection, leading in one third of the animals to septicaemia. Recovered from the blood at the death of the animal

the organisms were still non-encapsulated and avirulent for rabbits possessing leucocytes.

Since, in leucopenic animals, the avirulent pneumococcus behaves as a virulent pathogen, the transformation of the virulent, encapsulated form into the avirulent, non-encapsulated form entails no change in the bacteria which prevents their survival and free proliferation in the tissues if they are unmolested by phagocytes.

The local lesion produced by the non-encapsulated, avirulent pneumococcus in leucopenic animals exhibits the haemorrhages and necrosis of tissue familiar in the lesions produced by encapsulated, virulent pneumococci in the normal animal. The factor (polysaccharide capsule) responsible for virulence, therefore, is not necessary for the production of the characteristic type of tissue damage that virulent pneumococci produce. The power so to damage the tissues resides in the somatic rather than in the capsular portion of the bacterium, and the change in the organism resulting in the inability to produce a capsule does not alter the ability of the somatic portion to produce the characteristic tissue damage.

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BOOK REVIEWS

(These reviews represent the individual opinions of the reviewers and not necessarily those of the members of the Editorial Board of this Journal.)

Explorations in Personality. By HENRY A. MURRAY, M.D. 761 pp. $8.50. *Oxford University Press, New York, 1938.*

This book is the result of the coöperative effort of 28 different workers at the Harvard Psychological Clinic under the direction of Dr. Henry A. Murray. Working with a limited number of Harvard college students and young men from the ranks of the unemployed, the collaborators on this project devoted themselves to those special items necessary to the understanding of the present personality structure of the subjects by the use of historical method, special procedures for gaining additional material illuminating the biography, and by special tests of present capacities. The results of all these examinations were then pooled and an attempt was made to draw out of this effort a consistent procedure which might be used widely for the study and understanding of the human personality.

A prodigious amount of work went into this book. The authors compare their method very closely with that used in clinical medicine. Perhaps a better simile would have been clinical psychiatry in its broader applications. Yet the wealth of detail is overwhelming. The one case which is reported *in extenso* shows the method at work and its results.

Of special interest is the series of questionnaires designed to elicit qualitative personality distinctions. The outstanding short-cut in the method was the use of the Rorschach test for arriving at essentially many of the most important conclusions that have been more painstakingly developed by other means.

Not the least illuminating part of the whole case report is a retrospective account given by the subject some six months later, concerning the conference at which he was confronted by all the workers in the case. There is not enough of this sort of thing done in clinical practice. Usually too much is taken for granted concerning the patient's absorption of what is given him.

This is a rather formidable book not to be dealt with lightly. The terminology is rather difficult, and to this reviewer an unnecessarily large number of strange terms have been introduced, as, for example, *actone, press, blamavoidance, infavoidance, etc.* It appears that these were more or less neutral terms on which the various members of the working group, many holding biases of one sort or another, could agree.

There is an enormous amount of interesting detail in the book. The reviewer misses in it what he would consider to be the proper emphasis of a dynamic psychology, namely, the importance of the biographical method in which various

test situations serve only to give illustration to the theme which is carried by the story. We do things in a much more economical way in clinical psychiatry, and I should think that the perfection of the method which has been so resolutely attempted by the Harvard group also would finally yield much more economical treatment of the problem.

W. M.

Traité de Biocolloïdologie. By W. KOPACZEWSKI. Tome IV, *Etat colloïdal et biologie.* Fascicule VI, *Narcose.* 164 pp. 35 francs, paper bound. *Gauthier-Villars, Paris, 1936.*

This review assembles a voluminous literature (to 1933) on the subject of narcosis. The numerous theories are well summarized but not very critically analyzed. One gathers that there is no single mechanism that will explain narcosis but that the multitudinous vital activities of living cells can be reversibly interrupted at many points in long chains of physical and chemical reactions.

M. R.

Outline of Psychiatric Case-Study. By PAUL WILLIAM PREU, M.D. 140 pp. $1.85. *Paul B. Hoeber, New York, 1939.*

This is a handbook of psychiatric examination-method and record-taking as practiced at Yale University; but it is more than that. It contains much differential diagnostic material directly derived from the mental status examination and the record. It is a good book, containing in its small, compact size a wealth of information, the usefulness of which, I am sure, psychiatric experience generally will corroborate.

There is one deviation from the method in use in the Henry Phipps Psychiatric Clinic, namely, in the order of the case history. Long ago, we resorted to the practice of following the Complaint directly with the Present Illness, in order to give a rounded view of the current situation. This seems to us, after several years of experience, definitely preferable to the older system which is perpetuated in Dr. Preu's book and also in general medical practice, namely, of following the Complaint with the Family History, the Past History, and eventually coming to the Present Illness. However, this constitutes a minor matter in comparison with the general uniform excellence of the book.

W. M.

A Short Encyclopaedia for Nurses. By EVELYN C. PEARCE. 3rd ed. 686 pp. $3.50. *E. P. Dutton & Co., New York, 1939.*

The author's objectives in compiling this volume were to present graduate nurses with a ready reference book to meet their practical needs and to provide student nurses preparing for State Board Examinations with factual material in concise form, covering the usual subjects taught in the nursing curriculum.

An Index has been added to this edition, and the work has been brought up to date by the inclusion of new remedies used widely by physicians.

It is the reviewer's opinion that this book is of ample scope to suffice for general reference and might serve as the mainstay of the graduate nurse in the home, who is often compelled to solve daily problems and emergencies without aid or advice from others. In some instances the practices outlined are local, but for the most part the treatment and nursing care suggested are general and apply as well to the United States as to England.

It is doubtful whether students in Schools of Nursing in the United States would find this work, which is arranged alphabetically, as helpful in reviewing for State Board Examinations as some of the books published here which are classified according to the major subjects taught in the schools. In addition, students might find it difficult to discriminate between nursing care and treatment on a general level and that which represents local practice.

K. V. B.

Pulmonary Tuberculosis. A Synopsis. By JACOB SEGAL. 150 pp. Illus $2.75. *Oxford University Press, New York, 1939.*

This is a readable "capsule" exposition of present-day knowledge of the subject it presents.

In a small volume, the author has described clearly the etiology, pathogenesis, types, symptoms, usual complications, differential diagnosis and the treatment of pulmonary tuberculosis in a way to interest and to inform particularly those who desire a broad and superficial acquaintanceship with the disease. It should be interesting and helpful to social workers, to those active in the endeavors of Tuberculosis Associations, and to those who require no more than casual working information. To informed medical students and to physicians it is no more than an epitome of what should be to them familiar data.

The book omits a reference to Purified Tuberculin (P.P.D.) and contains a number of slightly inaccurate statements and some misspelled words. It is profusely illustrated but many of the illustrations lack sharp definition.

To those who seek merely an informative discussion of the topic, this book should be satisfying.

C. R. A.

Drug Addicts are Human Beings. By HENRY SMITH WILLIAMS, M.D. 273 pp. $2.50. *Shaw Publishing Co., Washington, D. C., 1938.*

The author claims that China and the United States are the only two modern countries where drug addiction is a real problem. He further claims that the problem has reached staggering dimensions in this country, owing to the operation of a set of rules formulated by the Commissioner of Internal Revenue, the Deputy Commissioner of the Narcotic Division of the Prohibition Unit, the Commissioner of Prohibition, ostensibly as enforcement procedures for the Harrison Act but in reality constituting a law unto themselves.

According to the author, the Harrison Act was designed as a revenue measure exclusively, and the matter of dispensing it was to be left entirely in the hands of

the medical profession. The code of rules mentioned above, however, according to the author, has changed the situation completely through the assumption that ambulatory treatment of narcotic addicts is inadvisable. The main effect of this has been to instil fear into the medical profession so that individual medical practitioners and even medical clinics established exclusively for the purpose, have been afraid to treat or have been enjoined from treating ambulatory addicts; and the addicts themselves, not being able to get the daily pittance of drugs in a legitimate fashion, have turned to the narcotic smuggler and peddler. The cruel thing in the entire procedure, as the author sees it, is that there is no provision for adequate hospital treatment for addicts. The author is certainly right when he calls attention to the fact that practically no general hospital will accept drug addicts for treatment, and special hospitals for the purpose are exceedingly rare.

The author concludes that these rules which have become, to all intents and purposes, law, are unconstitutional. He gives plenty of instances of physicians and medically sponsored clinics which have attempted to operate in defiance of the rules, to their own detriment. A Congressional investigation has been urged which might indeed throw some light upon the moot question of what is law and what is simply bureaucratic domination.

The author goes a little far when he draws an analogy between the treatment requirements for the diabetic and the myxedematous patient and those for the chronic drug addict who also seems to demand a daily ration of something which is missing from his system. This may indeed be the simplest way of viewing a bad situation. But the need for adequate hospital treatment facilities and the development of a general attitude toward the problem, which would favor treatment rather than attempts at reprisal, would seem to be the outstanding need from the social hygiene standpoint.

W. M.

A Textbook of Bacteriology. By THOMAS B. RICE. 2nd ed. revised. 563 pp. Illus. $5.00. *W. B. Saunders Co., Philadelphia, 1938.*

This book is exactly as claimed for it in the Preface, "a relatively simple text of such size that (students) may be expected to master it in one semester". "The inclusion of a bibliography has been deliberately rejected." It is stated that "Theoretical subjects should be handled in the lecture room or by additional reading". "Those who need more refined and detailed information will find abundant opportunity for study in other texts."

It is precisely for these reasons that I would not regard this textbook as adequate for students in Johns Hopkins Medical School. A student who masters one textbook is likely to be mastered by it. It is quite as important to teach doubt where doubt exists as to teach what, for the moment, may be regarded as well established fact. A textbook serves its most useful purpose by summarizing the literature and is incomplete without references to important original papers, old and new.

This book is well written and the material is well chosen. The Appendices contain valuable material not found in most other texts.

J. H. B.

Bacteria, The Smallest of Living Organisms. By FERDINAND COHN (1872). Translated by CHARLES S. DOLLEY (1881). Introduction by MORRIS C. LEIKIND. 44 pp. $1.00. *The Johns Hopkins Press, Baltimore, 1939.*

Cohn's paper is a classic of bacteriology, valuable for its appreciation of the entire field of bacteriology, interesting for the acute observations of the author with limited means of study available, and also for opinions which subsequent research has shown to be erroneous. It is an accurate picture of bacteriology in 1872, written at a time when pure bacteriology was about to be temporarily overshadowed by the more urgent applied bacteriology. The age of the microbe hunters was about to be ushered in by Koch's study of the anthrax bacillus and the attendant technique. Cohn realized the potentialities of bacteria, their wide distribution and their biological significance for good or evil, but the complexity of the world of "the smallest of living organisms" was yet to be revealed. In the meantime serology has been born, a two-edged instrument which not only helps to diagnose the pathogenic effects of bacteria but, even more, to reveal the systematic relationships of these organisms, pathogenic and non-pathogenic. Fifty years after the publication of Cohn's paper we find bacteriology increasingly concerned with the study of bacteria *per se* but with new and improved instruments. This perspective is necessary for appreciation of Cohn's paper.

It is inspiring to have this translation presented as the work of a medical student in 1881. The introduction to the reprint by Mr. Leikind leaves little for the reviewer and shows scientific as well as historical appreciation.

J. H. B.

BOOKS RECEIVED FOR REVIEW

Clinical Electrosurgery. By GUSTAVUS M. BLECH. 389 pp. Illus. $4.00. *Oxford University Press, New York, 1938.* (Oxford Medical Publications.)

Orthopedic Appliances. By HENRY H. JORDAN. 412 pp. Illus. $4.00. *Oxford University Press, New York. 1939.* (Oxford Medical Publications.)

Classic Descriptions of Disease. By RALPH H. MAJOR. 2nd ed. 727 pp. Illus. $5.50. *Charles C. Thomas, Springfield, Illinois, 1939.*

Drug Addicts are Human Beings. By HENRY SMITH WILLIAMS. 273 pp. $2.50. *Shaw Publishing Company, Washington, 1938.*

Treatment of Fractures. By CHARLES LOCKE SCUDDER. 11th ed. revised, 1208 pp. Illus. $12.00. *W. B. Saunders Company, Philadelphia, 1938.*

Trauma and Internal Disease. By FRANK W. SPICER. 593 pp. Illus. $7.00. *J. B. Lippincott Company, Philadelphia, 1939.*

Surgical Pathology of the Diseases of the Mouth and Jaws. By ARTHUR E. HERTZLER. 248 pp. Illus. $5.00. *J. B. Lippincott Company, Philadelphia, 1938.* (Hertzler's Monographs on Surgical Pathology.)

Diagnosis and Treatment of Diseases of the Thyroid. By JAMES H. MEANS and EDWARD P. RICHARDSON. 367 pp. Illus. $5.00. *Oxford University Press, New York, 1938.* (Reprinted from Oxford Monographs on Diagnosis and Treatment.)

Surgical Anatomy. By C. LATIMER CALLANDER. 2nd ed. entirely reset. 858 pp. Illus. $10.00. *W. B. Saunders Company, Philadelphia, 1939.*

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